VOLUME LXIX NUMBER 3

THE
BOTANICAL GAZETTE

MARCH 1920

SHORT CYCLE UROMYCES OF NORTH AMERICA[1]

G. R. BISBY

(WITH PLATE X)

The short cycle *Uromyces* may be segregated as a group by utilizing the criteria of life cycle and character of teliospores. Aside from any question of the validity of such bases for segregation, it is evident that it is a common practice thus to set apart this group, and that an opportunity is thereby afforded to consider relationships of such rusts to each other and to other rusts.

The short cycle *Uromyces* are of considerable interest, although as yet comparatively few species or even collections are recorded for North America. These rusts occur over a wide geographical range, however, and are parasitic upon widely separated families of hosts.

The writer has been privileged to examine the excellent collection of short cycle *Uromyces* in the herbarium of Dr. J. C. ARTHUR. This paper represents the results of the study of the group as made primarily for the *North American Flora*, but is presented separately in order to give notes and discussions not permissible in that publication.

[1] Abstract submitted before American Phytopathological Society at the New York meeting, and published in Phytopathology 7:74. 1917. Contribution from Botanical Department of Purdue University Agricultural Experiment Station.

Characters and relationships

The rusts considered in this paper are those which fulfil the following requirements:

Cycle of development includes only pycnia (sometimes) and telia, both subepidermal.

Pycnia deep-seated, globose or flask-shaped, with ostiolar filaments.

Telia erumpent, usually grouped; teliospores free, pediceled, 1-celled; wall firm, colored, smooth or variously sculptured; germination by a single promycelium from an apical pore.

Urediniospores normally absent but occasionally found in the telia.

The association of pycnia with telia has for some time been considered the criterion of short cycle rust (1, 4). The occurrence of definite aecia or uredinia (providing the evidence indicates that the aecia or uredinia belong with the telial stage present) suffices to exclude a specimen from the group under consideration. While some suspicion may be aroused by the presence of urediniospores, such spores occasionally occur in the telia. In the cases in which pycnia are only rarely or not at all produced, telia only being present, the arrangement and character of the telia usually may be utilized to indicate whether or not the specimen is short cycled; a grouping of the telia in definite circinating or crowded groups, or the occurrence of germination of the teliospores at or soon after maturity, usually means that such a specimen belongs with the group of rusts here treated. In certain cases, however, the telia are diffused, and other considerations must be brought to bear.

A study of the *Uromyces* forms of the rusts as represented in the Arthur herbarium and of the literature indicates that, in North America, only the 11 species described are at present known to belong in reality to short cycle forms.

DIETEL (10) pointed out that the percentage of endemic species of rusts is higher in proportion to the isolation of the geographical region; that *Uromyces* shows a higher percentage of species in warmer than in colder regions; and that in both the Old and the New World the number of species of *Uromyces* is about one-third that of the number of species of *Puccinia*. It is to be noted that, so far as known, 8 of the 11 short cycle species of *Uromyces* are endemic to North America, and only 1 of the 11 species occurs also in Europe.

These forms are more especially found in the warmer parts of the continent, just as all *Uromyces* seem to be more numerous in warmer regions. While in North America some three times as many species of *Puccinia* as of *Uromyces* exist, the relation of the forms when divided according to their life cycle is strikingly different; for about 140 short cycle species of *Puccinia* are known for North America, in contrast to these 11 species of *Uromyces*.

P. and H. SYDOW (**25**), in their monograph of *Uromyces*, described only the telial stage for 183 of the 504 species considered in that work. For only a very few of these, however, were pycnia described. When full information is in hand, a large number of the 183 forms will of course be found not to be short cycled. It appears, however, that a comparatively greater preponderance of these short cycle forms of *Uromyces* may be found in the more tropical regions. The observation of MAGNUS (**17**) and of FISCHER (**15**), that increased altitude results in shortened life cycles for the rusts, is somewhat borne out by the fact that certain short cycle *Uromyces* are limited to the Rocky Mountain region. The effect of altitude and temperature can be better noted with the more numerous short cycle species of *Puccinia*.

ORTON (**21**) has touched upon the relation of a group of rusts with a common life cycle, opsis forms of *Puccinia* (the genus *Allodus*), to other groups with different life cycles. Comparable relationships and correlations with other rusts are to be noted with the group of rusts considered in this paper; some attention is directed to these points with the discussion of the several species. The rust in its development is intimately dependent upon its host. FISCHER (**13**) in 1898 emphasized the similarity between the teliospores of certain short cycle rusts and of long cycle heteroecious rusts whose aecia occurred upon the host of the short cycle form. He considered that this similarity indicated a phylogenetic relationship between such rusts with different life cycles. DIETEL (**9**) considered that the Uredinales have probably developed during geological times along with their hosts. ARTHUR (**5**) has pointed out that the relationships of the rusts often reflect the relationships of the hosts upon which they occur. The writer (**6**) has also dealt somewhat with this point.

DIETEL (11) considered that *Uromyces* is the most primitive of the Pucciniaceae, both on account of the possession of 1-celled teliospores, and because it occurs upon such diverse families of monocotyledons and dicotyledons. Whether long cycle or short cycle rusts are more primitive is still a mooted question.

The existence of species of these rusts as lepto-forms or micro-forms, that is, whether or not the teliospores germinate upon maturity, while subject somewhat to seasonal variation, is a fairly constant and characteristic feature with each species.

Life history; cytology

The life cycle is simplified in a short cycle species to the extent that only telia, often with pycnia, are produced. The occasional occurrence of a few urediniospores in the telia is a phenomenon in common with other groups of rusts which ordinarily do not bear such spores.

FISCHER (12) first cultured a short cycle *Uromyces*. He sowed teliospores of *U. Cacaliae* (DC.) Unger upon *Adenostyles alpina* Kern, securing telia again without the intervention of any other spore stages. No trace of pycnia was found. In 1905 FISCHER (14) reported the culture of the short cycle species of *Uromyces* which occurs in Europe as well as in America, *U. Solidaginis*. He sowed teliospores upon *Solidago Virgaurea alpestris*, and in about 13 days noted the infection upon the leaves; telia were produced, but in no case were pycnia to be observed. While North American material of this species has not been cultured, it is supposed that similar conditions obtain here. SCHNEIDER (24) cultured *U. Scillarum* (Grev.) Wint., a short cycle form, and reported specialization as to hosts. The teliospores were found to be capable of germination, either at once or after a period of rest. No cultures of endemic North American short cycle *Uromyces* seem to have been reported. CARLETON (8), ARTHUR (3), and others, however, have reported cultures of some species of lepto-*Puccinia*. WILLE (27) recently found a sharp specialization of the lepto-form *Puccinia Arenariae* upon the different host genera attacked.

The evidence obtained from cultures indicates that similar conditions exist in the short cycle forms, both of *Uromyces* and *Puccinia*.

A greater specialization and fixity may exist with short cycle forms than with forms with long life cycles; of course fewer spore forms upon which variability may be manifested are present.

Pycnia may be produced, under certain conditions, in some of these short cycle species of *Uromyces* not yet known to produce pycnia. It is to be noted, however, that species in which the teliospores germinate at maturity, that is, lepto-forms, seldom produce pycnia. Teliospores cannot function directly as repeating spores, but in lepto-forms a comparatively rapid repetition is secured through the intervention of the basidiospores, which are produced immediately upon maturity of the teliospores.

Cytological work upon the short cycle rusts indicates that similar conditions obtain with the short cycle species, both of *Uromyces* and *Puccinia*. The work of SAPPIN-TROUFFY (**23**) upon the histology of the rusts included a study of the short cycle forms *Uromyces Ficariae* (Schum.) Lev. and *Puccinia malvacearum* Mont. His observations were corroborated and extended with the two rusts, among others, by BLACKMAN and FRASER (**7**). They found that the general vegetative mycelium of *Uromyces Ficariae* consists of uninucleate cells, some of the later vegetative, together with the sori-forming, mycelium being binucleate. They found similar conditions for *Puccinia malvacearum*, the binucleate condition evidently arising at several different points for each sorus, shortly before the sorus is formed. BLACKMAN and FRASER also observed that the short cycle forms *Puccinia Adoxae* Hedw.f. and *Uromyces Scillarum* (Grev.) Wint. had a binucleate rather than a uninucleate general vegetative mycelium, and suggested that it is "probable that in these two forms the conjugate condition is produced soon after infection by nuclear migration, or by cell fusion, between vegetative cells." OLIVE (**20**) discussed and figured sexual fusions near the base of the telium in a short cycle form, *Puccinia transformans* Ellis and Ev. Dealing with North American rusts, OLIVE (**19**) also reported that differing conditions as to the sporophytic and gametophytic generations occurred with certain short cycle *Puccinia* forms; while *Uromyces Rudbeckiae* Arth. and Holw. showed the anomalous extreme of possessing uninucleate cells through all the mycelium and sorus, even including the teliospores.

This phenomenon he was not able to explain fully. Other papers to be noted are those by WERTH and LUDWIGS (**26**), HOFFMAN (**16**), and MOREAU (**18**). A considerable summary of recent cytological work is presented by RAMSBOTTOM (**22**).

From this work it appears that the duration of the binucleate stage varies in different species of short cycle rusts, being brief, extended, or intermediate. Fusions between cells initiate this binucleate condition. Some life history problems, including the

TABLE I

HOST RELATIONSHIPS OF SHORT CYCLE *Uromyces*

Host	Species of Uromyces	Distribution	Mycelium	Common condition of germination	Pycnia
Liliaceae					
Erythronium...	U. heterodermus	Rockies	Local or rather diffuse	Micro	Present
Cassiaceae					
Bauhinia.....	U. bauhiniicola	S.W. Mexico	Rather diffuse	Micro	Present
Bauhinia.....	U. jamaicensis	Mexico; West Indies	Local	Micro	Present
Fabaceae					
Psoralea.....	U. abbreviatus	Pacific Coast	Local, becoming rather diffuse	Micro	Present
Euphorbiaceae					
Chamaesyce, Tithymalus	U. Tranzschelii	Western N.A.	Diffuse	Micro	Present
Primulaceae					
Primula.....	U. nevadensis	Western N.A.	Local or rather diffuse	Micro	Not known
Myrsinaceae					
Myrsines.....	U. Myrsines	Costa Rica; S.A.	Local	Micro	Not known
Carduaceae					
Solidago.....	U. Solidaginis	W.N.A.; Europe	Local	Micro	Not known
Anaphalis.....	U. amoenus	Western N.A.	Local	Micro	Not known
Rudbeckia.....	U. Rudbeckiae	Central N.A.	Local	Lepto	Not known
Bidens.....	U. Bidentis	Porto Rico; S.A.	Local	Lepto	Not known

relative importance and relation of cell and nuclear fusions, some relations in the formation of pycnia in short cycle forms, the presence of perennial mycelium, etc., appear not to have been fully determined.

Hosts

The range of hosts attacked by these North American short cycle species of *Uromyces* embraces both the monocotyledons and dicotyledons. The situation is shown in table I.

Foreign species of short cycle *Uromyces* fill in several families not represented here. The wide range of hosts attacked indicates that these rusts do not form a restricted group; one might expect

to find affiliations with other forms of rusts upon the same or similar hosts through the various families, and such is the case. Under the species *U. heterodermus* a considerable comparison with rusts from related hosts is made, suggesting that certain groups of hosts appear to harbor rusts characterized by various definite morphological characters.

The geographical distribution of North American short cycle species of *Uromyces* would indicate further that the mountainous and more tropical regions furnish the most favorable location for these forms. Only *U. Rudbeckiae* has a comparatively wide range, a range including the plains area.

Whether or not it is more than a coincidence that the absence of pycnia and the occurrence of lepto-germination are found on hosts higher in the evolutionary scale, the writer is not prepared to say.

Taxonomic

With the progress of critical studies of North American rusts, other short cycle forms will undoubtedly be separated out, and further evidence secured as to the fixity and definiteness of the life cycle in certain of these rusts. *Uromyces heterodermus*, for example, was long placed with *U. Erythronii*, a correlated form. It was found also that *U. Bidentis* was a short cycle form which resembled *U. bidenticola* (P. Henn.) Arth. so far as characters of teliospores are concerned. It is no doubt true that other short cycle forms have been collected and placed with correlated long cycle forms, although cultures are needed to determine the life cycle in certain cases.

The 11 species of *Uromyces* considered have several points of similarity, one of which is the fact that all possess teliospores with apices more or less thickened. In none of the species were paraphyses, stromata, isolated peridial cells, or other accompanying structures found in the telia.

Key

Teliospores verrucose.
  Teliospores long, up to 43–47 μ
    Wall up to 1.5 μ thick..... 1. *U. heterodermus*
    Wall up to 2.5 μ thick..... 6. *U. nevadensis*
  Teliospores short, up to 30 μ..... 5. *U. Tranzschelii*

Teliospores reticulate.
Spores 14–21 by 18–26 μ ..... 2. *U. bauhiniicola*
Spores 12–17 by 16–23 μ ..... 3. *U. jamaicensis*
Teliospores smooth.
Wall thin, 1–1.5 μ.
Spores narrow, 11–17 μ wide.
Spores 27–39 μ long ..... 7. *U. Myrsines*
Spores 19–32 μ long ..... 10. *U. Rudbeckiae*
Spores broad, 15–28 μ wide ..... 11. *U. Bidentis*
Wall thick, 1.5–3 μ.
Apex thickened 6–12 μ ..... 8. *U. Solidaginis*
Apex thickened 3–7 μ.
Spores 27–40 μ long ..... 4. *U. abbreviatus*
Spores 19–30 μ long ..... 9. *U. amoenus*

1. UROMYCES HETERODERMUS Sydow, Ann. Myc. **4**:29. 1906.

O. Pycnia amphigenous, not uncommon, gregarious in loose groups with the telia, 0.5–1.5 mm. across, inconspicuous, subepidermal, dark golden-brown, flattened globoid, 100–185 μ in diameter by 65–130 μ in height; ostiolar filaments few, loose, up to 65 μ long.

III. Telia amphigenous, numerous, scattered or in small groups, sometimes upon inconspicuous spots, roundish or oval, 0.3–2 mm. across, rather early naked, pulverulent, dark cinnamon-brown, surrounding epidermis noticeable; teliospores ellipsoid or broadly ellipsoid, 19–26×26–43 μ, rounded above, rounded or slightly narrowed below; wall dark golden-brown, 1.5 μ thick, thickened at the apex with a distinct hyaline papilla, 3–6 μ, coarsely verrucose above, with the markings often in longitudinal ridges, smoother below; pedicel hyaline, fragile, short.

ON LILIACEAE: *Erythronium grandiflorum* Pursh, Colorado, Montana, Utah, Washington, British Columbia; *E. montanum* S. Wats., Washington; *E. obtusatum* Goodding, Wyoming; *E. parviflorum* (Wats.) Goodding (*E. grandiflorum parviflorum* S. Wats.), Colorado, Montana, Oregon, Utah, Washington, Wyoming.

TYPE LOCALITY: Wasatch Mountains, Utah, on *Erythronium parviflorum*.

DISTRIBUTION: Rocky Mountain region from Colorado and Utah northward, and to the coast in Oregon.

EXSICCATI: Barth., Fungi Columb. *4694;* Barth., N.Am. Ured. *789, 1592, 1692;* Garrett, Fungi Utah. *118;* Ellis and Ev., Fungi Columb. *750*.

LITERATURE: SYDOW, Monog. Ured. **2**:270. 1910; SACCARDO, Syll. Fung. **21**:579. 1912.

This rust, previous to SYDOW's description in 1906, passed as *U. Erythronii* (DC.) Pass., a related European species possessing aecia. Thus ELLIS and EVERHART's Fungi Columbiani *750* was issued as *U. Erythronii*. The host of this collection is undoubtedly *Erythronium parviflorum;* earlier collections of this host were frequently considered, as in this case, to be *E. grandiflorum*.

This rust occurs upon the species of *Erythronium* found in the western part of North America. According to ENGLER (ENGLER and PRANTL, Nat. Pflanz. 2^5:60. 1888) species of *Erythronium* occur particularly in North America. He places the following genera together to constitute the section Liloideae-Tulipeae: *Lilium*, *Fritillaria*, *Erythronium*, *Tulipa*, *Lloydia*, and *Calochortus*. Several rusts occur upon these genera of hosts. For the sake of comparison, all such rusts are tabulated. To avoid a personal factor, the data are largely from the SYDOWS' *Monograph*, and any supplementary data obtained are added in brackets. Parentheses indicate a rather free translation. Some data are taken from a paper by REES (Amer. Jour. Bot. **4**:368–373. 1917), who also presents drawings which support the contention that the rusts on these hosts possess rather unusual morphological similarities.

Table II shows many points of similarity in these rusts. It is to be noted that practically all possess amphigenous, rounded or minute, pulverulent sori, with spores broadly ellipsoid, rather similar as to size, with the wall usually moderately thick, apex somewhat thickened with a papilla, pedicel hyaline and short; and especially, all possess, in a striking manner, surface markings usually striate or verrucose and arranged in rows. This unanimity in morphological characters would indicate that a closely and definitely related group of rusts occurs upon these related hosts. Correlations, more or less perfect, obtain throughout this group of rusts upon the Liloideae-Tulipeae, and are found to extend further through the Liliaceae. Figs. 1–6 illustrate, for comparison, the teliospores of three of these rusts.

2. UROMYCES BAUHINIICOLA Arth. BOT. GAZ. **39**:389. 1905.—*Telospora Bauhiniicola* Arth., Result. Sci. Congr. Bot. Vienne 346. 1906.

TABLE II

Characters of rusts of Liloideae-Tulipeae

Rust	Host genus	Source of data	Life cycle	Geographical distribution	Leaf surface bearing telia	Shape of telia	Character of telia
Uromyces heterodermus Sydow	Erythronium	This paper	O, III	N.A.	Amphigenous	Round or oval	Pulverulent
Uromyces aecidiiformis (Strauss) Rees	(Fritillaria?)	Rees	O, I, III	Europe	Amphigenous	Elongated	Pulverulent
Uromyces Lilii (Link) Fuckel	Lilium	Sydow	O, I, III	(N.A.?), Europe	Amphigenous	Round or oblong	Pulverulent
Uromyces Erythronii (DC.) Pass.	Erythronium	Sydow	O, I, III	Europe	Amphigenous	Roundish	Pulverulent
Uromyces mogianensis Bubak	Fritillaria	Sydow	O, I*, III	Asia, Europe, Africa	Amphigenous	Round or oblong	Pulverulent
Uromyces Miurae Sydow	Fritillaria	Rees	(O, I)? III	Japan, N.A.	Amphigenous	Round or ellipsoid	Pulverulent
Uromyces Fritillaria (Schlechtd.) Thüm	Fritillaria	Rees	O, I, III	Europe	Amphigenous	Elliptical	Pulverulent
Uromyces Tulipae Dietel	Tulipa	Sydow	I, III	Japan	Amphigenous	Minute	Pulverulent
Uromyces japonicus Sydow	Lilium	Sydow	II, III	Japan	(Amphigenous)	Round	Pulverulent
Uromyces Holwayi Lagerh	Lilium	Sydow	[O, I], II, III	N.A.	Amphigenous	Minute round	Pulverulent
Nigredo Lilii (Clint.) Arth	Lilium	Flora	O, I, II, III	N.A.	Amphigenous	.....	Pulvinate?
Puccinia Calochorti Peck	Calochortus	Sydow	(O), I, III	N.A.	Chiefly hypophyllous	Round	Pulverulent
Puccinia Prostii Mong	Tulipa	Sydow	III	Europe	Amphigenous	Oblong	[Pulverulent]
Puccinia Tulipae Schroet	Tulipa	Sydow	III	Europe	Chiefly hypophyllous	Minute	.....

Rust	General shape of teliospores	Size of teliospores	Wall in μ	Apex in μ	Apex character	Sculpturing	Pedicel
Uromyces heterodermus Sydow	Broad, ellipsoid	19–26×26–43	1–5	3–6	Hyaline papilla	Coarsely verrucose in rows	Hyaline, short
Uromyces aecidiiformis (Strauss) Rees	Broad, ellipsoid	23–27×31–35	About 3	[to 5]	Hyaline apiculus	Rugose in rows	Hyaline, short
Uromyces Lilii (Link) Fuckel	Broad, ellipsoid	22–30×28–44	2–3.5	[to 5]	Hyaline papilla	Rugose or verrucose in rows	Hyaline, short
Uromyces Erythronii (DC.) Pass.	(Broad, ellipsoid)	16–25×22–42	1.5–2	[to 6]	Hyaline papilla	Anastomose in rows	Hyaline, short
Uromyces mogianensis Bubak	Subglobose	26–32×26–38	3–6	.....	Hyaline papilla	Verrucose in rows	Hyaline, deciduous
Uromyces Miurae Sydow	Ellipsoid	14–23×24–35	1.5–2	to 5	Hyaline apiculus	Verrucose in rows	Hyaline, short
Uromyces Fritillaria (Schlechtd.) Thüm	Broad, ellipsoid	23–31×31–42	.....	to 5	Hyaline apiculus	Rugose in rows	Hyaline, short
Uromyces Tulipae Dietel	Broad, ellipsoid	20–28×26–40	.....	.....	Hyaline papilla	Reticulate-striolate	Hyaline, short
Uromyces japonicus Sydow	Ovate	20–28×28–46	1.5	.....	Hyaline papilla	Verrucose in rows	Hyaline, deciduous
Uromyces Holwayi Lagerh	Broad, ellipsoid	20–28×24–42	[2–2.5]	[5–7]	Hyaline papilla	Verrucose in rows	Hyaline, deciduous
Nigredo Lilii (Clint.) Arth	Broad, ellipsoid	18–25×29–39	2–2.5	5–7	Apiculus	Rugose in ridges	Hyaline, short
Puccinia Calochorti Peck	[Broad], ellipsoid	22–30×33–40	[1.5–2.5]	[3–5]	.....	Verrucose	Hyaline (short)
Puccinia Prostii Mong	(Broad), ellipsoid	34–40×54–66	[2–3]	[3]	(Aculeate)	Aculeate, strongly	[Hyaline] (short)
Puccinia Tulipae Schroet	Broad, ellipsoid	21–32×30–44	.....	.....	.....	Verrucose	Hyaline, short

* Bubak says O with III.

O. Pycnia epiphyllous, few, gregarious in small groups, usually opposite the telia, punctiform, subepidermal, brownish, flattened globose, 60–130 μ in diameter by 30–65 μ in height; ostiolar filaments compact, short.

III. Telia at first hypophyllous, becoming also somewhat epiphyllous, numerous, scattered or in small groups, roundish, small, 0.2–1 mm. across, early naked, pulverulent, chocolate-brown, surrounding epidermis inconspicuous; teliospores globoid or broadly ellipsoid, 14–21 by 18–26 μ, rounded at the ends; wall cinnamon or chestnut brown, thick, 2.5–4 μ, apex thicker, 4–7 μ, with a paler, broad umbo, finely reticulated; pedicel pale or colorless, often roughened below, rather fragile but sometimes two or three times as long as the spore.

On Cassiaceae: *Bauhinia Pringlei* S. Wats., Jalisco; *Bauhinia* sp., Guerrero.

Type locality: Guadalajara, Mexico, on *Bauhinia Pringlei*.

Distribution: Known only from Southwest Central Mexico.

Illustration: Ark. Bot. Stockh. 4: *pl. 1. fig. 9.*

Exsiccati: Barth., N.Am. Ured. *286.*

Literature: Vestergren, Ark. Bot. Stockh. 4:28–29. 1905; Sydow, Monog. Ured. 2:80, 81. 1909; Saccardo, Syll. Fung. 21:550–551. 1912.

3. Uromyces jamaicensis Vesterg. Ark. Bot. Stockh. 4:33. 1905.

O. Pycnia chiefly epiphyllous, gregarious in small groups with the telia, subepidermal, brownish, flattened, 60–100 μ in diameter by 45–70 μ in height; ostiolar filaments compact, hardly extending beyond the ostioles.

III. Telia amphigenous, numerous, gregarious in small groups or occurring singly, sometimes on small yellowish spots, roundish, small, 0.1–1 mm. across, early naked, pulverulent, chestnut-brown, surrounding epidermis noticeable; teliospores globoid, broadly ellipsoid or obovoid, 12–17×16–23 μ, rounded or slightly narrowed at the ends; wall cinnamon-brown, 1.5–2 μ (sometimes up to 3.5 μ) thick, thicker at the apex, up to 5 μ, with a lighter crater or cap-shaped crown, closely and finely reticulate, appearing verrucose under the lower powers of the microscope; pedicel pale, fragile, 4–15 μ long.

ON CASSIACEAE: *Bauhinia divaricata* L., Cuba, Guanajuoto; *B. Pauletia* Pers., Porto Rico; *B. porrecta* Sw., Jamaica.

TYPE LOCALITY: Constant Spring, Jamaica, on *Bauhinia* sp.

DISTRIBUTION: Mexico and the West Indies.

ILLUSTRATION: Ark. Bot. Stockh. **4**: *pl. 2. fig. 14.*

LITERATURE: SYDOW, Monog. Ured. **2**:84. 1909; SACCARDO, Syll. Fung. **21**:552–553. 1912.

This species may perhaps be distinguished from the preceding by the somewhat reduced length and breadth of the teliospores, the wall thickness often being less also. The differences described by VESTERGREN (*loc. cit.*) have not been found to hold entirely throughout the collections at the Arthur herbarium. Some differences, however, are still to be found between the two species of rust, and they are maintained as separate species, at least pending further collections.

VESTERGREN'S supposition that *Uromyces jamaicensis* is a micro-*Uromyces* has been corroborated by the discovery of pycnia associated with telia upon a Cuban specimen of *Bauhinia divaricata*. The specimen upon *B. porrecta* collected by THAXTER has not been seen, but VESTERGREN'S type collection has been examined.

VESTERGREN separated 17 species of *Uromyces* upon the host *Bauhinia*, for none of which aecia are known. Evident similarities are shown between the species as he described and figured them. *Uromyces* only are known to occur upon *Bauhinia*. Many species of *Bauhinia* occur in the tropics; related genera, as shown by ENGLER and PRANTL'S classification, are chiefly genera upon which rusts have not yet been found. The reticulate nature of the sculpturing upon the surface of the teliospores of these two species is minute, but evident under higher microscopic power. Figs. 7–10 illustrate the two species.

4. UROMYCES ABBREVIATUS Arth. Bull. Torr. Bot. Club **42**:587. 1915.

O. Pycnia hypophyllous, scattered among the telia, inconspicuous, subepidermal, deep seated, dark honey-yellow, globose or flattened globose, 115–200 μ in diameter by 95–140 μ in height; ostiolar filaments dense, often falling away, up to 60 μ in length.

III. Telia hypophyllous, rarely also epiphyllous, densely clustered, becoming scattered over considerable areas, roundish,

0.2–0.7 mm. across, early naked, pulverulent, chocolate-brown, surrounding epidermis at first evident, later often hidden by the loose spores; teliospores ellipsoid or irregularly obovoid, 21–26 × 27–40 μ (sometimes variable in size, and larger), rounded above, rounded or narrowed below; wall chestnut-brown, 1.5–3 μ thick, apex 3–5 μ thick, often with a slight umbo over the pore, smooth; pedicel colorless, delicate, fugacious, half as long as the spore or less.

On Fabaceae: *Psoralea physodes* Dougl., California, Washington, British Columbia; *P. Purshii* Vail, Nevada.

Type locality: Winnemucca, Nevada, on *Psoralea Purshii*.

Distribution: Pacific Coast region, west of the mountains, from British Columbia to California.

Exsiccati: Barth., N.Am. Ured. *1582;* D. Griff., W.Am. Fungi *390;* Barth., Fungi Columb. *4884*.

The type of this species is Griffith's West American Fungi *390*, which was issued as *Uromyces Psoraleae* Peck. *U. Psoraleae* possesses aecia, however. *U. abbreviatus*, while without aecia, and possessing pycnia with the telia, resembles *U. Psoraleae* in the telial stage, as indicated by Arthur in the notes with the original description. While *U. Psoraleae* extends to the Pacific Coast, it is more common in the Rocky Mountain region, and extends over the plains to the east of the mountains. *U. abbreviatus*, so far as known, is limited to the region west of the Rockies.

There is an unconnected *Aecidium* (*Aecidium onobrychidis* Burrill, Bull. Ill. State Lab. Nat. Hist. **2**:225. 1885) upon *Psoralea Onobrychis*, represented as far as known by the one collection by Seymour in Illinois, and distributed by Ellis and Everhart as North American Fungi *1826*. No other species of rust are reported for the genus *Psoralea*, and these species are only known in North America. Related hosts, as given by Engler and Prantl, except for the genus *Indigofera* in an adjoining section, are scarcely known to be attacked by rusts; no closely related rusts are evident upon related hosts.

While the type collection is from an altitude of about 5000 ft., other collections in the Arthur herbarium are from nearer the coast, at much less altitude, extending almost down to sea-level.

5. UROMYCES TRANZSCHELII Sydow; Tranzschel, Ann. Myc. 8:20. 1910.

O. Pycnia hypophyllous, scattered among the telia, or in groups, noticeable, subepidermal, dark yellow, globoid or flask-shaped, 100–145 μ in diameter by 75–130 μ in height; ostiolar filaments dense, agglutinated into a truncate column, 50–80 μ in height, 50–70 μ in diameter at the ostiole.

III. Telia hypophyllous, occasionally sparingly epiphyllous, numerous, evenly scattered over large areas, or sometimes in groups around the pycnia, roundish, 0.2–0.6 mm. across, early naked by a central pore, pulverulent, chestnut-brown, surrounding epidermis crateriform, conspicuous; teliospores globoid or ellipsoid, 15–22 × 19–30 μ, rounded at the ends, wall cinnamon-brown, 1.5–2.5 μ thick, apex 3–5 μ thick with a low, sub-hyaline apiculus, minutely verrucose, the markings often in irregular longitudinal lines; pedicel colorless, deciduous.

ON EUPHORBIACEAE: *Chamaesyce serpens* (H.B.K.) Small (*Euphorbia serpens* H.B.K.), California; *Tithymalus montanus* (Engelm.) Small (*Euphorbia montana* Engelm.), Colorado, New Mexico, Utah, Wyoming; *T. robustus* (Engelm.) Small (*Euphorbia montana robusta* Engelm.), Colorado, Utah, Wyoming; *Tithymalus* sp. (*Euphorbia Palmeri* Engelm.), Lower California.

TYPE LOCALITY: Colorado, on *Euphorbia montana*.

DISTRIBUTION: From Wyoming to New Mexico, California, and Lower California.

EXSICCATI: Barth., N.Am. Ured. *499;* Ellis and Ev., Fungi Columb. *1069;* Ellis and Ev., N.Am. Fungi *2230;* Garrett, Fungi Utah. *97*.

LITERATURE: TRANZSCHEL, Ann. Myc. 8:1–35. 1910; SYDOW, Monog. Ured. 2:171–172. 1910; SACCARDO, Syll. Fung. 21:560–561. 1912; DIETEL, Hedw. 28:185–187. 1889; ARTHUR, Bull. Torr. Bot. Club 45:152. 1918.

This rust passed as *Uromyces scutellatus* (Schrank.) Lev., a European species, until SYDOW's description in 1910. TRANZSCHEL pointed out that *U. Tranzschelii* is similar to *U. monspessulanus* Tranz.; indeed, other similarities to various Euphorbiaceous rusts are evident. In his study of the autoecious rusts upon *Euphorbia*, TRANZSCHEL stated that most European autoecious species with telia from diffused mycelium had passed as two species, *Uromyces scutellatus* or *U. excavatus;* he divided such forms into some 12 species, and found a total of 27 autoecious species of *Uromyces*

upon hosts belonging to the various sections of the genus *Euphorbia*. That these species are related is evidenced by the fact that many had passed under one name; furthermore, many similarities are to be noted from TRANZSCHEL's descriptions. For example, all but one species are listed as having verrucose or striolate teliospore walls. A table showing characters in a manner similar to those tabulated under *U. heterodermus* would be illuminating as indicating relationships between *U. Tranzschelii* and other species of rust upon related hosts. The writer considered it sufficient, however, to call attention to TRANZSCHEL'S work as indicating relationships. Certain heteroecious species with aecia upon *Euphorbia* likewise show resemblances to *U. Tranzschelii*.

Both ELLIS and EVERHART'S Fungi Columbiani *1069* and North American Fungi *2230* were issued as *U. scutellatus*, while GARRETT'S Fungi Utahensis *97* was issued as *U. andinus* P. Magn., a related South American rust.

The range of *U. Tranzschelii* begins at about the western limit of the range of the related species *U. proeminens* (DC.) Pass., and continues westward to the Pacific Coast. Range conditions comparable with those of *U. abbreviatus* are thus shown, and neither of these short cycle forms necessarily occurs at high altitudes.

TRANZSCHEL (*loc. cit.*, p. 20) considered the rust upon *Euphorbia Palmeri* to be different, apparently another species. The specimen studied by the writer is not considered different from other specimens of *U. Tranzschelii*.

DIETEL (*loc. cit.*) commented upon ELLIS and EVERHART'S North American Fungi *2230*, especially concerning the relationship of an *Aecidium* upon the same host distributed as no. *2215* of the same exsiccati. It is true that *Aecidium Tithymali* Arth. occurs upon the same hosts, sometimes upon the same leaves, as *Uromyces Tranzschelii*. The situation in regard to this *Aecidium Tithymali* is uncertain. Germination tests show that it is a true *Aecidium* and not an *Endophyllum*. Its alternate host, however, has not been found. ARTHUR (*loc. cit.*) has discussed this *Aecidium* and its possible relation to *U. Tranzschelii*.

6. UROMYCES NEVADENSIS Hark. Bull. Calif. Acad. Sci. 1:36. 1884.—*Caeomurus nevadensis* Kuntze, Rev. Gen. 3^3:450. 1898.

O. Pycnia unknown.

III. Telia amphigenous, circinating in groups 2–5 mm. across, or somewhat scattered, round or oval, 0.2–1.0 mm. across, early naked, pulvinate, becoming somewhat pulverulent, chestnut-brown, ruptured epidermis conspicuous; teliospores oblong, oblong-obovoid, or ellipsoid, 19–27×29–47 μ, rounded at the apex, rounded or narrowed toward the base; wall cinnamon-brown, lighter or colorless at the apex, moderately thick, 1.5–2.5 μ, thickened at the apex, 5–7 μ, moderately and rather finely verrucose; pedicel colorless, fragile.

On Primulaceae: *Primula suffrutescens* Gray, Nevada.
Type locality: Lake Tahoe, Nevada, on *P. suffrutescens*.
Distribution: Known only from the type locality.
Literature: Magnus, Ber. Deutsch. Bot. Gesells. **18**:451–459. 1900.
Illustration: Magnus, *loc. cit. pl. 16. figs. 16–19.*

The writer is considerably indebted to the members of the botanical staff at the Purdue Station for the preceding. Arthur in a letter states that "a careful study of this species seems to leave little doubt that it is a distinctly American species and a short cycle one. This was the conclusion reached by Magnus in 1900." The one collection known was made by Harkness, and a specimen has been studied by the writer.

7. Uromyces Myrsines Diet. Hedwigia **36**:26. 1897.

O. Pycnia unknown.

III. Telia hypophyllous, crowded upon reddish or brownish spots 2–10 mm. in diameter, margin of spots usually elevated, roundish, 0.1–0.2 mm. in diameter, often confluent, early naked, pulvinate, light chocolate-brown, ruptured epidermis inconspicuous; teliospores oblong or oblong-ellipsoid, 13–16×27–39 μ, rounded or acute above, narrow below; wall pale golden-brown, rather thin, 1–1.5 μ, thickened at the apex, 3–8 μ, smooth; pedicel colorless, short.

On Myrsinaceae: *Ardisia compressa* H.B.K., Costa Rica.
Type locality: Rio de Janeiro, Brazil, on *Myrsine* sp.
Distribution: Costa Rica; also in South America.
Literature: Arthur, Mycologia **10**:124, 1918; Sydow, Monog. Ured. **2**:46. 1909.

This rust was known only from South America before its discovery by HOLWAY in one locality in Costa Rica. South American specimens have been distributed by E. ULE, Herbarium Brasiliense no. *2136*. ARTHUR suggests that *U. marginatus* Bomm. and Rouss may be a synonym. SYDOW gives *U. Rhapaneae* Henn. and *U. Usterianus* Diet. as synonyms. While SYDOW was probably right, it has been impossible to examine specimens of these two collections.

8. UROMYCES SOLIDAGINIS (Sommerf.) Niessl, Verh. Natur. Ver. Brunn **10**:163. 1872.—*Caeoma Solidaginis* Sommerf. Suppl. Fl. Lapp. 234. 1826; *Caeomurus Solidaginia* Kuntze, Rev. Gen. 3^3:450. 1898; *Telospora Solidaginia* Arth., Result. Sci. Congr. Bot. Vienne 346. 1906.

O. Pycnia not found; probably wanting.

III. Telia hypophyllous, sometimes also petiolicolous or caulicolous, crowded and often confluent in orbicular groups upon the leaves, or in elongated groups upon the petioles or stems, 2–10 mm. across, upon yellowish spots, roundish, small, 0.3–0.7 mm. across, early naked, compact, pulvinate, chocolate-brown, surrounding epidermis noticeable; teliospores obovate or ellipsoid, 17–25 × 24–33 μ, narrowed or rounded at the ends; wall chestnut-brown, 1.5–3 μ thick, much thicker at the apex, 6–12 μ, smooth; pedicel nearly colorless, about as long as the spore.

ON CARDUACEAE: *Solidago polyphylla* Rydb., Colorado; *S. serotina* Ait., Montana, Washington, Wyoming.

TYPE LOCALITY: Nordland, Sweden, on *Solidago virgaurea*.

DISTRIBUTION: Colorado to Montana and Washington, also in Europe and Asia.

ILLUSTRATIONS: Archiv. Naturw. Land. Bohmen **13**: *fig. 12;* Beitr. Krypt. Schweiz 2^2: *fig. 44*.

EXSICCATI: D. Griff., W.Am. Fungi *361*; Ellis and Ev., N.Am. Fungi *2883*.

LITERATURE: COOKE, Grev. **5**:152. 1877; WINTER, in Rab. Krypt. Fl. 1^1:141. 1881; SACCARDO, Syll. Fung. **7**:566. 1888; FISCHER, Beitr. Krypt. Schweiz 2^2:59, 543. 1904; FISCHER, Ber. Schw. Bot. Gesells. **15**:(1–2). 1905; HARIOT, Les Ured. 216. 1908; SYDOW, Mónog. Ured. **2**:10. 1909.

This is the one species of *Uromyces* included in this paper which is not endemic to the Americas. FISCHER (1905) reported cultures of this rust. He also (1898) pointed out the correlation between this species and *U. Junci* (Desmaz.) Tulasne, which bears aecia

upon hosts related to *Solidago*. The range of both *U. Solidaginis* and *U. Junci* in North America is similar, both occurring in the western part.

Uromyces Junci-effusi Sydow resembles *U. Solidaginis* in the telial stage; the aecial connection is not known for this form. Curiously not *Puccinia Solidaginis* Peck, but *P. Asteris* Duby (both are short-cycled) shows a correlation with *Uromyces Solidaginis*. Of the short cycle species of *Puccinia* upon *Solidago* in America, one, *P. Virgauriae* (DC.) Lib., is more eastern, possesses stromata, and has thin-walled teliospores. *P. Solidaginis*, although a western form, has very large teliospores. *P. Asteris*, however, is very similar to *Uromyces Solidaginis* in gross and microscopic characters, except in the possession of 2-celled teliospores. *Puccinia Asteris* is a more common rust, and while rare west of the Rockies, is found over most of North America. Figs. 19–22 illustrate *U. Solidaginis* from America and Europe and *P. Asteris*.

COOKE (*loc. cit.*) reported *Uromyces Solidaginis* from Maine. Collections from Eastern North America are not at hand; further doubt may be attached to COOKE's reported collection from the fact that he states that the spores are reticulated. GRIFFITH's West American Fungi *361*, although issued as *Puccinia Solidaginis*, is in reality *Uromyces Solidaginis*.

9. UROMYCES AMOENUS Sydow, Ann. Myc. 4:28. 1906.

O. Pycnia unknown.

III. Telia hypophyllous, densely grouped and often confluent on circular purplish spots, 2–8 mm. across, the margin of the spots yellow, roundish, small, 0.2–0.7 mm. across, early naked, compact pulvinate, dark chestnut-brown, covered by the tomentose pubescence of the host, ruptured epidermis inconspicuous; teliospores globoid, obovoid, or ellipsoid, 16–23 × 20–30 μ, usually rounded above and narrowed below; wall dark golden-brown or cinnamon-brown, moderately thick, 1.5–2.5 μ, apex thicker, 4–7 μ, smooth; pedicel pale yellowish, up to the length of the spore.

ON CARDUACEAE: *Anaphalis margaritacea occidentalis* Greene, Oregon; *A. subalpina* (A. Gray) Rydb. (*A. margaritacea subalpina* A. Gray), Idaho, Oregon, Washington, Wyoming, British Columbia.

TYPE LOCALITY: Washington, on "*Gnaphalium* (*Anaphalis*) *margaritacea*."

DISTRIBUTION: Wyoming to British Columbia and Oregon.

EXSICCATI: Ellis and Ev., Fungi Columbiani *1795;* Barth., N.Am. Ured. *1385, 1584.*

LITERATURE: SYDOW, Monog. Ured. 2:4. 1909; SACCARDO, Syll. Fung. 21:570. 1912.

Several collections of this rust are in the Arthur herbarium. Although the hosts of some collections are labeled *Anaphalis margaritacea*, it would appear that the name *A. subalpina* should be used for almost all collections in hand (compare COULTER and NELSON, New Manual of the Botany of the Central Rocky Mountains, p. 537).

ELLIS and EVERHART'S Fungi Columbiani *1795* was issued as *Uromyces Gnaphalii* Ellis and Ev., but is *U. amoenus*. *U. Gnaphalii* has been found to be a synonym of *U. intricatus* Cooke.

10. UROMYCES RUDBECKIAE Arth. and Holw. Bull. Iowa Agric. Coll. 1884. 154. 1885.—*Caeomurus Rudbeckiae* Kuntze, Rev. Gen. 3^3:450. 1898; *Telospora Rudbeckiae* Arth., Result. Sci. Congr. Bot. Vienne 346. 1906.

O. Pycnia unknown.

III. Telia hypophyllous, occasionally also epiphyllous, densely gregarious upon brownish spots, paler below, 1–10 mm. across, rather circinate, small, 0.2–0.8 mm. in diameter, early naked, compact, pulvinate, cinnamon-brown, soon cinereous from germination, surrounding epidermis not noticeable; teliospores ellipsoid, obovoid, or pyriform, 11–17×19–32 μ, rounded, acute, or obtuse at the apex, narrowed below; wall yellowish or very pale chestnut-brown, thin, 1 μ, apex thicker, 5–8 μ, smooth; pedicel hyaline, twice as long as the spore or less.

ON CARDUACEAE: *Rudbeckia laciniata* L. (*R. ampla* A. Nels.), Colorado, Illinois, Indiana, Iowa, Michigan, Minnesota, Missouri, Montana, Nebraska, New Mexico, North Dakota, Pennsylvania, Wisconsin, Wyoming, Ontario; *Rudbeckia* sp., Texas.

TYPE LOCALITY: Decorah, Iowa, on *Rudbeckia laciniata*.

DISTRIBUTION: Ontario and Pennsylvania to Montana and Texas.

ILLUSTRATION: Arth. and Holw. Ured. Exs. Ic. 1: *pl. 1. fig. 1.*

EXSICCATI: Arth. and Holway, Ured. Exs. Ic. 1: Barth., Fungi Columb. *4394;* Barth., N.Am. Ured. *299, 1099, 1397;* Brenckle, Fungi Dak. *274;* Ellis and Ev., Fungi Columb. *2097;* Ellis, N.Am. Fungi *1439;* Rab.-Wint., Fungi Eur. *3412;* Sydow, Ured. *1305, 1962.*

LITERATURE: BURRILL, Bull. Ill. State Lab. Nat. Hist. **2**:163. 1885; SACCARDO, Syll. Fung. **7**:581. 1888; ARTHUR and HOLWAY, Bull. Lab. Nat. Hist. State Univ. Iowa **3**:44. 1895; SYDOW, Monog. Ured. **2**:7–8. 1909.

Uromyces Rudbeckiae has been collected more frequently than any North American species of the group. Its range embodies the greater part of the plains area, and extends to the Rocky Mountains.

DIETEL (Ann. Myc. **8**:305. 1910) considered *Uromyces Komerovii* Bubak on *Solidago Virgaurea* identical with *U. Rudbeckiae*. No specimens of the former have been seen, although a collection on *Solidago Virgaurea* in the herbarium has not been found to differ from *U. Solidaginis*.

The only other rust found upon *Rudbeckia* is *Aecidium Compositarum* Auct., recently found to belong with *U. perigynius* Halsted (Mycol. **9**:307), a connection suspected from the fact that the telial stage of *U. Rudbeckiae* is similar to the telial stage of *U. perigynius*. A type of correlation which has frequently been of service in indicating alternate stages of heteroecious rusts is thus evidenced. The cytological work upon this species is noted earlier in this paper.

11. UROMYCES BIDENTIS Lagerh. Bull. Soc. Myc. Fr. **11**:213, 1895.—*Caeomurus Bidentis* Kuntze, Rev. Gen. 3^3:449. 1898; *Uromyces densus* Arth. Mycologia **7**:196. 1915.

O. Pycnia unknown.

III. Telia hypophyllous, numerous, in small circinating groups on roundish, discolored spots, 1–4 mm. across, sometimes confluent, roundish or oval, 0.1–1 mm. across, the central sorus larger, surrounded by smaller ones, early naked, compact, pulvinate, dull cinnamon-brown, becoming waxy-cinereous from germination, surrounding epidermis inconspicuous; teliospores obovoid or oblong, 15–28×30–45 μ, rounded or narrowed above, narrowed below; wall pale golden or cinnamon-brown, thin, 1 μ, thicker above, 4–9 μ, smooth; pedicel hyaline, once or twice the length of the spore or less.

ON CARDUACEAE: *Bidens leucantha* (L) Willd., Porto Rico; *B. pilosa* L., Porto Rico; *Bidens* sp., Costa Rica.

TYPE LOCALITY: Ecuador, South America, on *Bidens andicola*.

DISTRIBUTION: Porto Rico and Central America; also in South America.

The SYDOWS (Monog. Ured. **1**:3. 1909) misapplied LAGERHEIM'S name to the species with uredinia, now called *Uromyces bidenticola* (P. Henn.) Arth. The situation in regard to these two rusts is discussed by ARTHUR (Mycologia **9**:71. 1917), and he also (Mycologia **10**:127. 1918) suggests that it is possible that a fixity of life cycle may not occur in these *Bidens* rusts. *U. Bidentis* is correlated with *U. Bidenticola*, differing only in the life cycle and in the characters of the telia, which are coalescent and thickened into cushions in *U. Bidentis*. Specimens are at hand also from South America; LAGERHEIM'S collection from the type locality has been examined.

Puccinia Bidentis Diet. and Holw., BOT. GAZ. **24**:32. 1897, collected by HOLWAY in Mexico, apparently is not a correlated species.

EXCLUDED SPECIES

UROMYCES HYALINUS Peck, BOT. GAZ. **3**:34. 1878.—*U. Sophorae* Peck, Bull. Torr. Bot. Club **12**:35. 1885; *Caeomurus hyalinus* Kuntze, Rev. Gen. 3^3:450. 1898; *Telospora hyalina* Arth., Result. Sci. Congr. Bot. Vienne 346. 1906.

LITERATURE: SACCARDO, Syll. Fung. **7**:581. 582. 1888; HARIOT, Revue Mycol. **14**:21. 1892; SYDOW, Monog. Ured. **2**:128. 1909.

This rust, first described upon *Sophora sericea* from Colorado, and made the type of the genus *Telospora*, has been found to possess uredinia. OLIVE, in his paper on intermingling of perennial sporophytic and gametophytic generations, etc. (Ann. Myc. **11**:309. 1913), mentions that ARTHUR has called attention "to the fact that *Uromyces Sophorae* seems to possess a similar habit [that is, an intermingling of mycelia] to the perennial rusts under discussion.". In any event, the presence of uredinia, in some cases at least, suffices to exclude this species from the short cycle forms.

UROMYCES PAVONIAE Arth., Bull. Torr. Bot. Club **31**:1. 1914.—*Telospora Pavoniae* Arth., Result. Sci. Congr. Bot. Vienne 346. 1906.

LITERATURE: SACCARDO, Syll. Fung. **17**:250. 1905; SYDOW, Monog. Ured. **1**:59. 1909.

This rust, described upon *Malache scabra* B. Vogel (*Pavonia racemosa* L.) from Porto Rico and Jamaica, belongs with *Puccinia*

heterospora Berk. and Curt. An examination of the material shows that a very few 2-celled teliospores are present. *P. heterospora*, upon related Malvaceous plants, is characterized by the preponderance of 1-celled mesospores such as those upon *Pavonia*.

ARTHUR (Mycologia **9**:80. 1917) has given a brief discussion of the situation here. *Uromyces pictus* Thuem. upon *Abutilon* was also found to possess a few 2-celled teliospores and was placed with *Puccinia heterospora* by SYDOW (Monog. Ured. **2**:58 and 356. 1910).

UROMYCES MONTANA Arth., BOT. GAZ. **39**:386. 1905.—*Telospora montana* Arth., Result. Sci. Congr. Bot. Vienne 346. 1906.

The type collection of this species possessed also aecia, which were at the time considered to belong with *Uromyces Lupini* B. and C. Subsequent collections in Guatemala by KELLERMAN and HOLWAY, however, show the same association of aecia and telia; furthermore, these aeciospores are larger and thicker walled than the aeciospores of *U. Lupini*. The grouped arrangement of the telia and the thin walls of the teliospores and their germination at maturity indicate a short cycle form, but nevertheless it is considered probable that the aecia go with the telia. *U. elatus* Syd., also upon *Lupinus*, shows the same situation as regards association of aecia with telia resembling those of a short cycle form. I am indebted to Dr. MAINS of Purdue for work upon this species.

UROMYCES CUPANIAE Arth., Mem. Torr. Bot. Club **17**:131. 1918.—*Uredo cristata* Speg., Anal. Soc. Ci. Argent **17**:119. 1884.

This rust, although short-cycled, is excluded from this group, since, as noted by ARTHUR, it has marked affinities with other groups of rusts rather than with the group herein treated.

Conclusions

Eleven species of *Uromyces* possessing only telia and pycnia, or telia alone, are now considered to be present in North America. These are found especially in the higher and warmer portions of the continent, and occur upon 7 widely separated host families. While these rusts form a group agreeing as to life cycle and as to the 1-celled character of the teliospores, it is not considered that phylogenetic interrelationship is thereby shown, morphological evidence indicating rather that the relationship of a

species of these rusts is found in other rusts (of various life cycles and with 1 or 2-celled teliospores) upon the same or related hosts. Indeed, as indicated under *Uromyces heterodermus*, a group of hosts may bear a number of rusts of various life cycles, belonging to *Puccinia* and *Uromyces*, widely distributed geographically, yet all showing a certain unanimity of morphological characters, especially in the telial stage.

The writer wishes to express his keen appreciation to Dr. J. C. ARTHUR for suggesting this paper and for much help, and also to Professor JACKSON for many suggestions and constructive criticism. To the other workers in the laboratory at Purdue University he is likewise greatly indebted.

UNIVERSITY OF MINNESOTA
ST. PAUL, MINN.

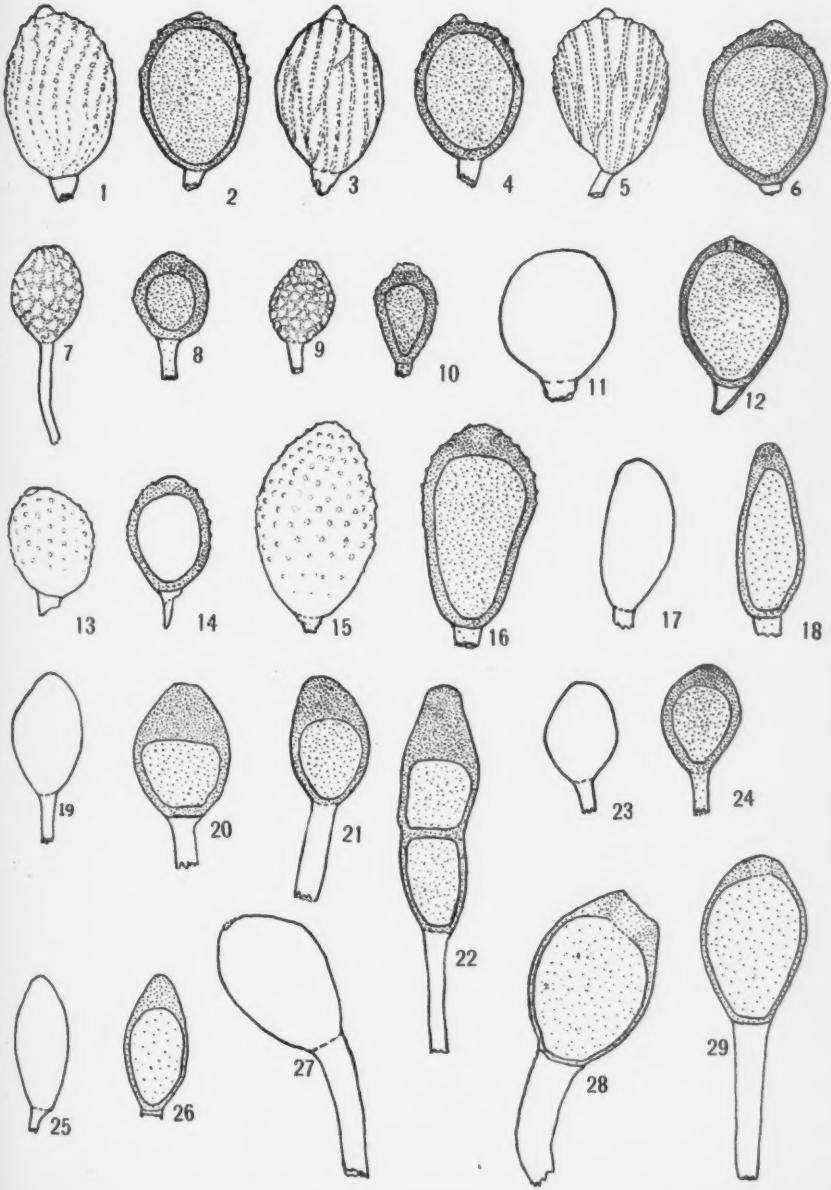
LITERATURE CITED

1. ARTHUR, J. C., Taxonomic importance of the spermogonium. Bull. Torr. Bot. Club **31**:113–123. 1904.
2. ———, Result. Sci. Congr. Bot. Vienne. 346. 1906.
3. ———, Cultures of Uredineae in 1905. Jour. Myc. **12**:20–21. 1906.
4. ———, Uredinales, North American Flora **7**:130. 1907.
5. ———, North American rose rusts. Torreya **9**:21–28. 1909.
6. BISBY, G. R., The Uredinales found upon the Onagraceae. Amer. Jour. Bot. **3**:527–561. 1916.
7. BLACKMAN, V. H., and FRASER, H. C. I., Further studies on the sexuality of the Uredineae. Ann. Botany **20**:35–48. 1906.
8. CARLETON, M. A., Investigations of rusts. Bull. no. 63, Bur. Pl. Ind. 1–29. 1904.
9. DIETEL, P., Betrachtungen über die Verteilung der Uredineen auf ihren Nährpflanzen. Centralb. Bakt. u. Par. **12²**:218–234. 1904.
Über die morphologische Bewertung der gleichnamigen Sporenformen in verschiedenen Gattungen der Uredineen. Hedw. **48**:118–125. 1908.
10. ———, Einige Bemerkungen zur geographischen Verbreitung der Arten aus den Gattungen *Uromyces* und *Puccinia*. Ann. Myc. **9**:160–165. 1911.
11. ———, Über die auf Leguminosen lebenden Rostpilze und die Verwandtschaftsverhältnisse der Gattungen der Pucciniaceen. Ann. Myc. **1**:3–14. 1903; see also Ann. Myc. **10**:205–213. 1912.
12. FISCHER, ED., Entwickelungsgeschichtliche Untersuchungen über Rostpilze. Beitr. Krypt. Schweiz **1**:7–8. 1898.

13. FISCHER, ED., Recherches sur les Urédinées suisses. Berne, 1898; original not seen; abs. by HARIOT, PAUL, Les Urédinées, pp. 83–85. Paris. 1908.
14. ———, Fortsetzung der Entwickelungsgeschichtlichen Untersuchungen über Rostpilze. Ber. Schweiz. Bot. Gesells. **15**:1–13. 1905.
15. ———, Über den Einfluss des Alpinen Standortes auf den Entwickelungsgang der Urédinéen. Verh. Schweiz Nat. Ges. **88**:47. 1906.
16. HOFFMAN, A. W. HANS, Zur Entwickelungsgeschichte von *Endophyllum Sempervivi*. Centralb. Bakt. Par. **32²**:137–158. 1911.
17. MAGNUS, P., Über die auf Compositen auftretenden *Puccinia* mit Teleutosporen von Typus der *Puccinia Hieracii* nebst Andeutungen über den Zusammenhang ihrer specifischen mit ihrer vertical Verbreitung. Ber. Deutsch. Bot. Gesells. **11**:453–464. 1893; see also Hedw. Beibl. **39**:147–150. 1900.
18. MOREAU, MME F., Les phenomenes de la sexualite chez les Urédinées. These de la Faculti des sciences l'Universite de Paris. Ser. 779, no. 1563, Portiers. 1914: pp. 142. *pls. 14*. 1914; original not seen; review by FISCHER, ED., Zeitsch. Bot. **8**:360–362. 1916.
19. OLIVE, E. W., The nuclear condition in certain short-cycled rusts (abstract). Science **33**:194. 1911.
20. ———, Sexual cell fusions and vegetative nuclear divisions in the rusts. Ann. Botany **22**:331–360. *pl. 22*. 1908.
21. ORTON, C. R., North American species of *Allodus*. Mem. N.Y. Bot. Gard. **6**:175–208. 1916.
22. RAMSBOTTOM, J., Recent published results on the cytology of fungus reproduction. Trans. Brit. Myc. Soc. **5**:271–303. 1916.
23. SAPPIN-TROUFFY, P., Recherches histologiques sur la famille des Urédinées. Le Botaniste **5**:59–244. 1896.
24. SCHNEIDER, WERNER, Zur Biologie der Liliaceen bewohnenden Uredineen. Centralb. Bakt. u. Par. **32²**:452. 1912.
25. SYDOW, P and H., Monograph. Ured. **2**:1–296. 1909–1910.
26. WERTH, E., and LUDWIGS, K., Zur Sporenbildung bei Rost- und Brandpilzen. (*Ustilago antherarum* Fries und *Puccinia malvacearum* Mont.) Ber. Deutsch. Bot. Gesells. **30**:522–528. 1912.
27. WILLE, F., Zur Biologie von *Puccinia Arenariae* (Schum.) Wint. Ber. Deutsch. Bot. Gesells. **33**:91–95. 1915.

EXPLANATION OF PLATE X

All figures were drawn at the level of the stage with the aid of a camera lucida, with Leitz 1/12 oil immersion and ocular 4. They are here reduced one-third, so that the magnification is 667 diameters. Surface markings, where present, are indicated, and the stippling on the optical cross-section diagrams



BISBY on UROMYCES

represents to some degree the comparative darkness of color of the spore walls. In most cases the drawings of short cycle *Uromyces* were made from type material.

FIGS. 1, 2.—Surface and optical cross-section, respectively, of teliospores of *Uromyces heterodermus* Syd., from type material, on *Erythronium parviflorum*, Wasatch Mountains, Salt Lake County, Utah; A. O. Garrett, Fungi Utahensis *118*.

FIGS. 3, 4.—*Uromyces Erythronii* (DC.) Pass. on *Erythronium dens-canis*, Bohemia. Sydow, Ured. *1505;* species correlated with preceding.

FIGS. 5, 6.—*Uromyces Holwayi* Lagerh. on *Lilium columbianum*, Washington. Barth., N.A. Ured. *1387;* compare two preceding species.

FIGS. 7, 8.—*Uromyces bauhiniicola* Arth., from type material on *Bauhinia Pringlei*, Guadalajara, Mexico.

FIGS. 9, 10.—*Uromyces jamaicensis* Vesterg., from type material, on *Bauhinia* sp., Constant Spring, Jamaica.

FIGS. 11, 12.—*Uromyces abbreviatus* Arth., from type material, on *Psorlea Purshii*, Winnemucca, Nevada. Griffith, W.Am. Fungi *390*.

FIGS. 13, 14.—*Uromyces Tranzschelii* Sydow, from type material, on *Tithymalus* (*Euphorbia*) *montana*, Fossil Creek, Colorado. Ellis and Everhart, Fungi Columbiana *1069*.

FIGS. 15, 16.—*Uromyces nevadensis* Hark., from type material, on *Primula suffrutescens*, near Lake Tahoe, Nevada.

FIGS. 17, 18.—*Uromyces Myrsines* Diet. on *Ardisia compressa*, south of Cartago, Costa Rica.

FIGS. 19, 20.—*Uromyces Solidaginis* (Sommerf.) Niessl. on *Solidago serotina*, Helena, Montana. Ellis and Everhart, N.A. Fungi *2883*.

FIG. 21.—*Uromyces Solidaginis* on *Solidago virgaurea*, Sweden. Sydow, Ured. *2406*.

FIG. 22.—*Puccinia Asteris* Duby on *Aster adscedens*, Salt Lake County, Utah, illustrating correlation with *Uromyces Solidaginis*.

FIGS. 23, 24.—*Uromyces amoenus* Sydow, from type material, on *Anaphalis subalpina*, Paradise Valley, Mount Tacoma, Washington.

FIGS. 25, 26.—*Uromyces Rudbeckiae* Arth. and Holw., from type material, on *Rudbeckia laciniata*, Decorah, Iowa.

FIGS. 27, 29.—*Uromyces Bidentis* Lagerh. on *Bidens pilosa*, Ponce, Porto Rico, from type material of *Uromyces densus* Arth.

FIG. 28.—*Uromyces Bidentis* from material from type locality on *Bidens andicola*, Ecuador.

EFFECT OF SALTS UPON OXIDASE ACTIVITY OF APPLE BARK

CONTRIBUTIONS FROM THE HULL BOTANICAL LABORATORY 263

D. H. ROSE, HENRY R. KRAYBILL, AND R. C. ROSE

(WITH FIVE FIGURES)

Introduction

In an earlier paper (21) one of the authors showed that there is a marked difference in the action of the salts of the alkali metals upon the fire-holding capacity of tobacco, even when the salts have similar anions. For instance, the carbonates of potassium, rubidium, and caesium promote the combustion of tobacco to a very much greater extent than the carbonates of sodium and lithium. The chlorides of sodium, lithium, and potassium retard the combustion, but the chloride of potassium is not nearly so effective as the chloride of sodium or lithium. In general, the salts of potassium, rubidium, and caesium are much more favorable to combustion than those of sodium and lithium.

It has been known for a long time that potassium is an essential element for the higher plants. Numerous attempts have been made to replace potassium by sodium, and, while apparently sodium can fulfil some of the functions of potassium, attempts to replace potassium entirely by sodium have been unsuccessful. The fact that potassium seems to have such a marked property of promoting the combustion of tobacco, and sodium does not, suggests that this particular property of potassium may have a relation to certain functions in the plant, which cannot be fulfilled by sodium. These facts suggested that a study of the effect of the alkali salts upon oxidase activity might be of interest. The work reported in this paper was done in 1917. More extended studies were planned, but, since it has been impossible to carry them out completely at the present time, it seemed wise to report the results obtained.

Historical

BERTRAND (5) was the first investigator to point out that the salts of metals influence oxidase activity. He showed that manganese salts greatly increase the oxidase activity of preparations from alfalfa. GESSARD (15) found that the formation of melanin from tyrosin is increased in the presence of salts of the metals. BACH (4) substantiated GESSARD'S results, and showed that aluminum sulphate, salts of calcium, magnesium, manganese, and zinc increase melanin formation from tyrosin. The effect of the salts is to increase the further change of the oxidation product rather than to activate the taking up of oxygen. Aluminum salts hasten the formation of purpurogallin from the yellow oxidation product of the action of oxidase upon pyrogallol. BACH believed that the oxidation process is retarded by the accumulation of the primary oxidation products, and that the salts act to release them. WOLFF (32) found that the oxidation of tyrosin by tyrosinase from *Russula delica* is increased by the addition of small quantities of disodiumphosphate. PORODKO (26), ASO (3), ALSBERG (2), and EWART (11) have shown that salts of the metals give a blue color with guaiacum. PORODKO and EWART believed these salts to be inorganic oxidases. PORODKO pointed out that those metals which form salts of two degrees of oxidation are particularly active. ALSBERG, and also EWART, confirmed PORODKO'S observation and found that the chlorides of many of the metals give a blue color with guaiacum. ALSBERG attributed an important part in the reaction to the chlorine. EWART further found that the chlorides, nitrates, and sulphates of the same metal are not necessarily equally powerful in their action. Apparently the chlorides are more active than the sulphates. Various salts were found to act as sensitizers or retardants to oxidase activity. Potassium chloride, potassium iodide, potassium bromide, and potassium fluoride retard or even prevent the browning of pounded apple pulp.

Numerous investigators have shown that oxidase activity is affected by changes in reaction of the medium. BERTRAND (6) showed that the action upon guaiacol of laccase from *Rhus succedanea* is inhibited by 0.002 M concentration of sulphuric acid.

WOLFF found tyrosinase from *Russula delica* most active in a solution neutral to phenolphthalein, and ABDERHALDEN and GUGGENHEIM (1) found that tyrosinase is destroyed by 0.016 N hydrochloric acid, and greatly retarded by 0.016 N sodium hydroxide. ROSE (28) showed that the decrease in oxidase activity, as observed in the Bunzell apparatus, is due to an increase in the hydrogen ion concentration of the medium. REED (27) found oxidase activity in potatoes and apples inhibited even by low hydrogen ion concentrations; and likewise BUNZELL (9) found the action of oxidase retarded with increasing hydrogen ion concentrations.

Methods

All but one of the experiments described in this paper were made with portions of apple bark which had been dried at 35–40° C. for 2–3 hours, ground fine enough to pass through a 40-mesh wire sieve, and stored air dry in zinc-capped Mason jars. One experiment was made with solutions of precipitated oxidase separated from aqueous extracts of healthy bark and of diseased bark by the addition of about 10 volumes of alcohol. In order to obtain the precipitated oxidase, 2 gm. of bark were allowed to stand in a beaker with 10 cc. of water and 5 drops of toluol for 1 hour. The extract was then squeezed out through moist cheesecloth on coarse filter paper. The beaker was washed with five 1 cc. portions of water and the filter paper finally with two more. There was then added 50 cc. of 95 per cent alcohol to the filtrate (concentration of alcohol about 70 per cent) and the whole allowed to stand for 10 minutes. The flocculent precipitate which had formed was collected on a hard filter by gentle suction with a filter pump. There was then added 150 cc. more alcohol to the filtrate (concentration of alcohol now about 90 per cent) and the whole allowed to stand for 1 hour, since precipitation was slow, before this second fraction was collected on the filter with the first. The precipitate was dissolved in water and used immediately, as described later.

The stock solutions of all of the salts tested were made to a concentration of 0.5 N. Potassium chloride, manganese chloride, ferrous chloride, and ferric chloride were used also in the additional

concentrations of 0.1 N and 0.01 N. Since there was always 5 cc. of water in the apparatus, the final concentration of the salt, there was 0.1 N for 0.5 N solutions and 0.02 and 0.002 N for 0.1 N and 0.01 N solutions used.

Oxidation was measured in centimeters of mercury rise by means of the simplified BUNZELL apparatus (**8**). The shaking machine was run at the rate of 106 complete excursions per minute. All experiments were run for 3 hours, readings being taken every 15 minutes, and a final reading the following morning. When bark was used, the mixtures in the apparatus contained 0.1 gm. of bark, 1 cc. of salt solution, and 4 cc. of 1 per cent pyrogallol solution or salt and pyrogallol with bark omitted, the second combination serving as a control on the first. Preliminary experiments had shown that during the time in which these experiments were run the autoxidation of the pyrogallol was usually not more than the equivalent of 0.15 cm. mercury rise. In the experiment with precipitated oxidase, the precipitate from 2 gm. of bark was dissolved in 20 cc. of water, and 2 cc. of the solution, containing the dissolved precipitate obtained from 0.2 gm. of bark, were put in each apparatus, together with the usual amount of pyrogallol and water. All tests were run in duplicate. Two controls were run with each experiment, one containing only water, the other bark (or oxidase solution), pyrogallol, and water, but without the addition of salts.

The figures for P_H given in table VII were obtained by means of the apparatus described by ROSE (**28**).

Discussion

The chlorides in general retard oxidase activity. The chlorides of potassium, sodium, and lithium depress markedly the oxidation of pyrogallol by bark (table I). Similar results were obtained with all the other chlorides tested, except ferrous chloride (table VI). Ferrous chloride in 0.1 N concentration with bark and pyrogallol showed 1.79 cm. mercury rise, and with pyrogallol alone 1.45 cm., compared with the control of pyrogallol and bark as 1.00 cm. Since ferrous chloride is readily oxidized when exposed to the air, it is quite probable that the oxygen absorption for the most part represents that absorbed in the oxidation of ferrous chloride.

Results

The results of the experiments are shown in tables I–VII and figs. 1–5.

TABLE I

EFFECT OF 0.1 N KCl, NaCl, AND LiCl ON OXIDATION OF PYROGALLOL BY POWDERED HEALTHY APPLE BARK; TEMPERATURE 23.2–23.6° C.*

TIME OF READING	NO BARK			BARK			
	KCl	NaCl	LiCl	Check	KCl	NaCl	LiCl
May 21							
12.30.....	0.00	0.00	0.00	0.00	0.00	0.00	0.00
12.45.....	0.03	0.13	0.00	0.03	0.06	0.07	0.03
1.00.....	0.03	0.13	0.00	0.08	0.05	0.11	0.05
1.15.....	0.08	0.20	0.00	0.23	0.15	0.18	0.15
1.30.....	0.05	0.17	0.02	0.25	0.15	0.18	0.15
1.45.....	0.05	0.13	0.00	0.33	0.15	0.21	0.15
2.00.....	0.07	0.18	0.00	0.38	0.15	0.24	0.16
2.15.....	0.08	0.19	0.05	0.43	0.19	0.27	0.21
2.30.....	0.08	0.19	0.04	0.45	0.19	0.31	0.25
2.45.....	0.07	0.17	0.05	0.45	0.25	0.30	0.25
3.00.....	0.05	0.16	0.05	0.50	0.23	0.32	0.26
3.15.....	0.09	0.19	0.06	0.65	0.26	0.36	0.29
3.30.....	0.10	0.20	0.05	0.68	0.28	0.35	0.32
May 22							
8.40.....	0.00	0.00	0.00	1.25	0.80	0.74	0.77

* In tables I–V manometer readings in cm. of mercury corrected against an apparatus containing only water.

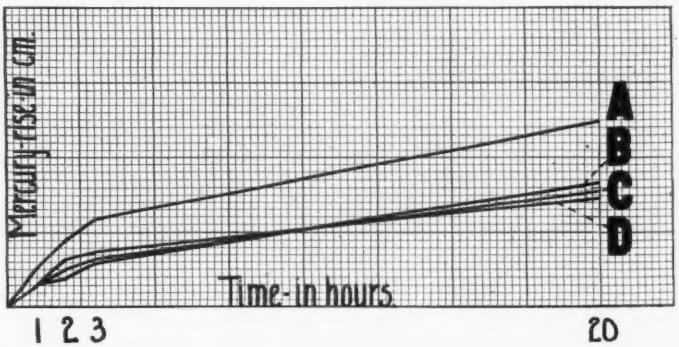


FIG. 1.—Effect of KCl, NaCl, and LiCl on oxidation of pyrogallol by powdered healthy apple bark: A, control (bark and pyrogallol); B, KCl+bark and pyrogallol; C, NaCl+bark and pyrogallol; D, LiCl+bark and pyrogallol.

TABLE II

EFFECT OF 0.10 N ALKALI CARBONATES ON OXIDATION OF PYROGALLOL BY POWDERED HEALTHY APPLE BARK; TEMPERATURE 29.3–30.0° C.

TIME OF READING	NO BARK			BARK			
	K_2CO_3	Na_2CO_3	Li_2CO_3	Check	K_2CO_3	Na_2CO_3	Li_2CO_3
June 13							
1.30.....	0.00	0.00	0.00	0.00	0.00	0.00	0.00
1.45.....	0.55	0.50	0.54	0.14	0.08	0.10	0.13
2.00.....	1.35	1.13	1.19	0.19	0.48	0.47	0.48
2.15.....	1.75	1.55	1.64	0.24	0.75	0.85	0.82
2.30.....	2.10	1.78	1.92	0.34	0.93	1.02	1.02
2.45.....	2.40	2.05	2.12	0.44	1.16	1.27	1.23
3.00.....	2.50	2.15	2.24	0.49	1.33	1.42	1.33
3.15.....	2.60	2.28	2.34	0.54	1.44	1.52	1.50
3.30.....	2.90	2.41	2.55	0.63	1.63	1.76	1.74
3.45.....	3.00	2.47	2.57	0.68	1.79	1.86	1.85
4.00.....	3.05	2.54	2.64	0.74	1.87	1.97	1.93
4.15.....	3.08	2.60	2.69	0.79	1.93	2.02	1.99
4.30.....	3.17	2.68	2.75	0.86	2.08	2.19	2.22
June 14							
8.30.....	3.17	2.63	2.74	1.29	2.70	2.61	2.85

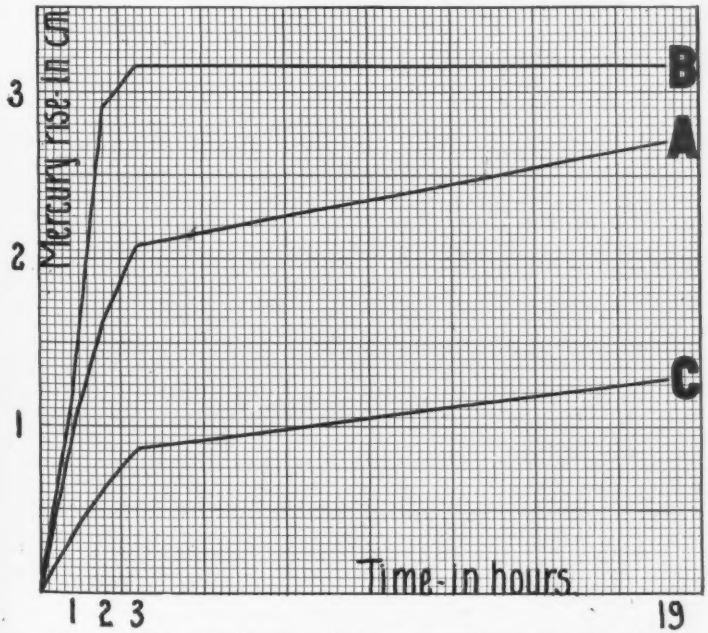


FIG. 2.—Effect of K_2CO_3 on oxidation of pyrogallol, with and without bark (healthy): A, K_2CO_3+bark and pyrogallol; B, K_2CO_3+pyrogallol; C, control (bark and pyrogallol).

TABLE III

EFFECT OF 0.10 N KCl AND K_2CO_3 ON OXIDATION OF PYROGALLOL BY POWDERED DISEASED APPLE BARK; TEMPERATURE 27.8–29.0° C.

TIME OF READING	NO BARK		BARK		
	K_2CO_3	KCl	Check	K_2CO_3	KCl
March 10					
10.00	0.00	0.00	0.00	0.00	0.00
10.15	0.68	−0.05	0.13	0.46	0.16
10.30	1.24	0.00	0.30	0.90	0.33
10.45	1.65	0.00	0.50	1.25	0.38
11.00	1.98	−0.08	0.65	1.50	0.48
11.15	2.25	−0.03	0.72	1.72	0.60
11.30	2.38	−0.03	0.85	1.93	0.69
11.45	2.52	0.00	0.99	2.09	0.82
12.00	2.65	−0.05	1.04	2.22	0.88
12.15	2.75	0.00	1.15	2.35	0.95
12.30	2.78	−0.05	1.18	2.39	0.95
12.45	2.85	−0.08	1.25	2.55	1.00
1.00	2.99	−0.05	1.38	2.65	1.10
March 11					
9.45	3.53	−0.10	2.20	3.73	1.73

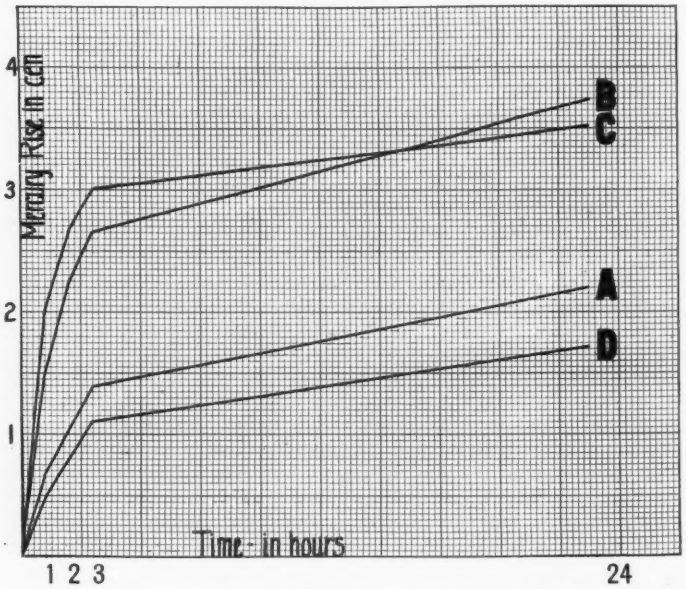


FIG. 3.—Effect of KCl and K_2CO_3 on oxidation of pyrogallol with and without bark (diseased): A, control (bark and pyrogallol); B, K_2CO_3+bark and pyrogallol; C, K_2CO_3+pyrogallol; D, KCl+bark and pyrogallol (KCl+pyrogallol gave no oxidation).

TABLE IV

EFFECT OF 0.10 N POTASSIUM TARTRATE, SODIUM OXALATE, AND $Ca(NO_3)_2$ ON OXIDATION OF PYROGALLOL BY POWDERED HEALTHY APPLE BARK; TEMPERATURE 29.2–30.2° C.

TIME OF READING	NO BARK			BARK			
	Potassium tartrate	Sodium oxalate	$Ca(NO_3)_2$	Check	Potassium tartrate	Sodium oxalate	$Ca(NO_3)_2$
June 22							
1.30	0.00	0.00	0.00	0.00	0.00	0.00	0.00
1.45	0.11	0.08	0.18	0.10	0.14	0.20	0.08
2.00	.....	.....	.....	0.25	0.28	0.25	0.20
2.15	0.25	0.10	0.20	0.30	0.36	0.35	0.23
2.30	.....	.....	.....	0.38	0.48	0.46	0.30
2.45	0.35	0.19	0.20	0.43	0.58	0.55	0.35
3.00	.....	.....	.....	0.55	0.64	0.64	0.40
3.15	0.48	0.31	0.28	0.58	0.76	0.78	0.50
3.30	.....	.....	.....	0.70	0.98	0.80	0.55
3.45	.....	.....	.....	0.73	0.95	0.96	0.59
4.00	0.68	0.38	0.35	0.80	1.03	1.00	0.60
4.15	.....	.....	.....	0.90	1.13	1.10	0.73
4.30	0.78	0.35	0.38	0.90	1.15	1.13	0.70
June 23							
8.20	0.95	0.68	0.23	1.20	1.53	1.60	0.98

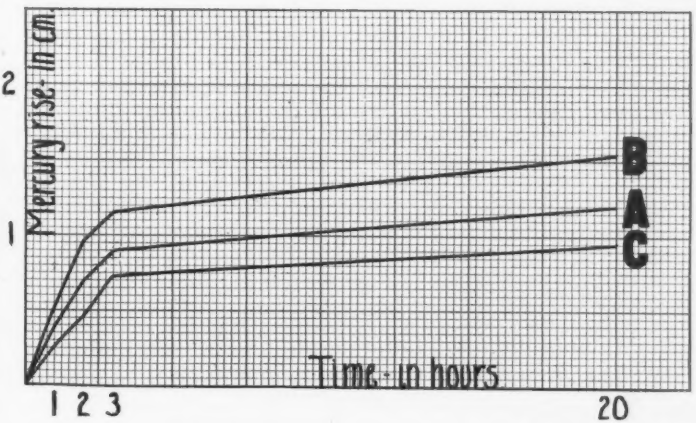


FIG. 4.—Effect of potassium tartrate on oxidation of pyrogallol with and without bark (healthy): A, control (bark and pyrogallol); B, potassium tartrate+bark and pyrogallol; C, potassium tartrate+pyrogallol.

TABLE V

EFFECT OF 0.10 N $MnCl_2$ AND K_2SO_4 ON OXIDATION OF PYROGALLOL BY PRECIPITATED OXIDASE FROM BOTH HEALTHY AND DISEASED APPLE BARK; TEMPERATURE 29.5–30.2° C.

TIME OF READING	HEALTHY			DISEASED		
	Check	$MnCl_2$	K_2SO_4	Check	$MnCl_2$	K_2SO_4
June 21						
1.45	0.00	0.00	0.00	0.00	0.00	0.00
2.00	0.07	0.08	0.11	0.17	0.15	0.15
2.15	0.08	0.10	0.21	0.37	0.29	0.30
2.30	0.08	0.13	0.23	0.42	0.21	0.33
2.45	0.08	0.13	0.27	0.48	0.25	0.43
3.00	0.08	0.10	0.25	0.50	0.23	0.48
3.15	0.15	0.11	0.28	0.56	0.24	0.54
3.30	0.15	0.08	0.30	0.65	0.26	0.58
3.45	0.18	0.09	0.35	0.70	0.29	0.63
4.00	0.20	0.08	0.34	0.79	0.31	0.69
4.15	0.20	0.08	0.37	0.87	0.34	0.78
4.30	0.23	0.09	0.38	0.88	0.35	0.78
4.45	0.28	0.18	0.43	0.98	0.40	0.93
June 22						
8.00	0.53	0.28	0.58	1.24	0.63	1.28

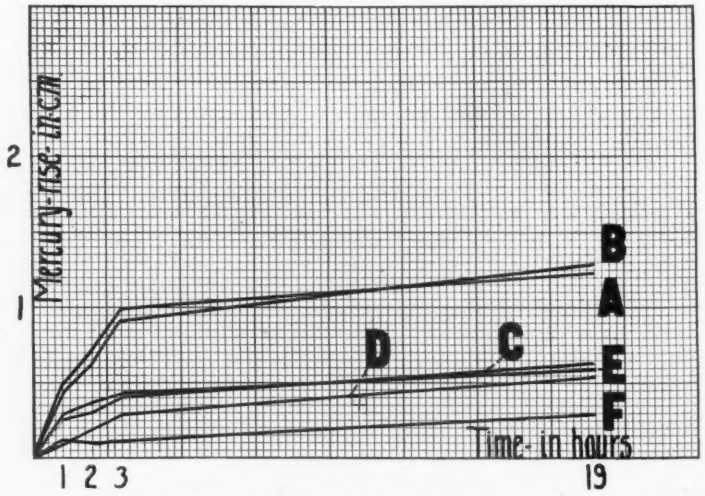


FIG. 5.—Effect of $MnCl_2$ and K_2SO_4 on the oxidation of pyrogallol on precipitated oxidase from both healthy and diseased bark: A (diseased), K_2SO_4+bark and pyrogallol; B (diseased), control (bark and pyrogallol); C (diseased), $MnCl_2$+bark and pyrogallol; D (healthy), K_2SO_4+bark and pyrogallol; E (healthy), control (bark and pyrogallol); F (healthy), $MCnl_2$+bark and pyrogallol.

TABLE VI

SUMMARY OF EFFECT OF ALL SALTS TESTED ON OXIDATION OF PYROGALLOL WITH AND WITHOUT BARK; ALL FIGURES CALCULATED TO BASIS OF 1 CM. MERCURY RISE FOR CHECK.

Salt		Check	Cl		SO_4		NO_3		CO_3		PO_4		H_2PO_4		Tartrate		Oxalate		Acetate		Citrate	
			With	Without	With	Without	With	Without	With	Without	With	Without	With	Without	With	Without	With	Without	With	Without	With	Without
K	0.1 N...	1.00	0.63	0.00	1.07	0.08	0.99	0.07	2.09	2.46	1.85	1.90	0.96	0.16	1.27	0.76	1.16	.....	.....	.....	.....	.....
K	0.02 N..	1.00	1.00	.....	.....	.....	.....	.....	.....	.....	.....	.....	1.04	.....	.....	.....	.....	.....	.....	.....	.....	.....
K	0.002 N.	1.00	1.00	.....	.....	.....	.....	.....	.....	.....	.....	.....	1.07	.....	.....	.....	.....	.....	.....	.....	.....	.....
Na	0.1 N...	1.00	0.59	0.00	1.06	0.06	0.98	0.09	2.02	2.11	.....	.....	.....	.....	1.14	0.26	1.33	0.55	1.55	1.84	2.06	2.41
Li	0.1 N...	1.00	0.61	0.00	1.05	0.10	.....	.....	2.20	2.19	.....	.....	.....	.....	.....	.....	.....	.....	1.37	1.28	1.92	2.09
Cs	0.1 N...	1.00	0.66	0.10	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
NH_4	0.1 N...	1.00	0.69	0.17	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
Ca	0.1 N...	1.00	0.57	0.15	.....	.....	0.80	0.15	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
Mg	0.1 N...	1.00	.....	.....	.....	.....	0.94	0.12	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
Mn	0.1 N...	1.00	0.54	0.10	1.04	0.02	0.76	0.04	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
Mn	0.02 N..	1.00	1.00	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
Mn	0.002 N.	1.00	1.15	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
Ba	0.1 N...	1.00	.....	.....	.....	.....	0.84	0.10	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
Fe'''	0.1 N...	1.00	1.79	1.45	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
Fe'''	0.02 N..	1.00	1.12	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
Fe'''	0.002 N.	1.00	1.10	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
Fe''	0.1 N...	1.00	0.24	0.05	.....	.....	0.31	0.24	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
Fe''	0.02 N..	1.00	0.55	.....	.....	.....	0.55	.....	.....	.....	.....	.....	.....	.....	.....	.....	0.45	0.06	.....	.....	.....	.....
Fe''	0.002 N.	1.00	0.86	.....	.....	.....	1.00	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
Mn	0.1 N...	1.00	0.50	.....	1.03	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
K	0.1 N...	1.00	0.52	.....	1.09	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....

TABLE VII

RELATION OF OXIDATION TO INITIAL P_H OF MIXTURES*

SALT	CHECK		Cl		SO_4		NO_3		CO_3		H_2PO_4		TARTRATE		OXALATE		ACETATE		CITRATE	
	Oxidation	P_H	Oxidation	P_H	Oxidation	P_H	Oxidation	P_H	Oxidation	P_H	Oxidation	P_H	Oxidation	P_H	Oxidation	P_H	Oxidation	P_H	Oxidation	P_H
K	1.00	5.15	0.63	5.19	1.07	5.13	0.99	5.14	....	.....	0.96	4.47	1.27	6.00	1.16	5.77	.....	5.72	.....	5.65
Na	.....	.....	0.59	5.17	.....	.....	.....	.....	.....	.....	.....	.....	1.14	.....	1.33	.....	1.55	6.43	2.06	6.02
Li	.....	.....	.....	.....	1.05	5.06	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	1.37	5.97	1.92	6.19
NH_4	.....	.....	0.69	4.85	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
Ca	.....	.....	0.57	4.79	.....	.....	0.80	4.74	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
Mg	.....	.....	.....	.....	.....	.....	0.94	4.62	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
Mn	.....	.....	0.54	4.48	1.04	4.50	0.76	4.43	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
Ba	.....	.....	.....	.....	.....	.....	0.84	4.78	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
Fe	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
Fe''	.....	.....	0.24	1.00	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....

* Concentration of salts 0.10 N.

This view is substantiated by the fact that when the concentration of ferrous chloride is reduced, oxygen absorption is reduced proportionally (table VI). If we subtract 1.45 cm. (mercury rise for pyrogallol and ferrous chloride) from 1.79 cm. (mercury rise for bark, pyrogallol, and ferrous chloride), we have 0.34 cm. for the oxidase activity of the bark in the presence of the ferrous chloride as compared with 1.00 cm. for the oxidase activity of bark and pyrogallol in the absence of ferrous chloride. Apparently ferrous chloride retards oxidase activity just as the other chlorides do, and the increased absorption of oxygen in the presence of ferrous chloride is due to the action of ferrous chloride itself in absorbing oxygen. Oxidation is increased by 0.002 N manganese chloride. This is in accord with the results of BERTRAND (**5**) and others. In a concentration of 0.1 N it inhibits oxidation just as do the other chlorides.

The use of precipitated oxidase shows that chlorides have a depressing effect on oxidation, even under conditions which eliminate many of the substances present in the bark powder. No investigation has been made of the effect of these substances on the reaction, but they probably complicate it.

The results with the chlorides are in accord with the work of EWART, who found that dilute solutions of potassium chloride and sodium chloride prevent the browning of slices of apples. EWART'S further conclusion, however, that the chlorides act as sensitizers to oxidation, or ALSBERG'S idea that chlorine plays an important part in the bluing of guaiacum by the chlorides of metals, are scarcely borne out by our observations that chlorides in general depress oxidase activity. It should be noted, however, that the results of those investigators were based upon color reactions, while ours were based upon oxygen absorption.

It is interesting to note that the chlorides which retard the combustion of tobacco at high temperatures have a similar effect in depressing oxidase activity. KRAYBILL (**21**) has suggested that the chlorides may have a negative catalytic action in the case of the combustion of tobacco. It would be interesting to know how the chlorides affect other oxidation processes.

The depressing effect of chlorides on oxidase activity is in contrast with their action on other enzymatic processes. Thus NASSE (25), KÜBEL (22), COLE (10), WOHLGEMUTH (31), LISBONNE (23), HAWKINS (18), and others have found that chlorides increase the diastatic power of various preparations of diastase. NASSE, however, found that under certain conditions sodium chloride retarded diastatic activity, and later HAWKINS showed that sodium chloride and potassium chloride in certain dilute concentrations (M/128–M/512) retard diastatic activity. It would have been better if the effect of the chlorides upon oxidase activity had been determined in a greater number of concentrations, and it will be well in the future to do so in studying this problem. The effect of salts upon lipase activity is also of interest in this connection. LOEVENHART and PEIRCE (24), GERBER (14), TERROINE (30), HAMSIK (16), FALK (12), and others found that the chlorides of various alkalies and alkaline earths retard lipase activity. TERROINE found that the concentration of the salts which he studied determined the nature of their influence. BUCHNER, BUCHNER, and HAHN (7) found that the chlorides of sodium, calcium, barium, and ammonium inhibit the fermentation of cane sugar or glucose in the presence of pressed yeast.

The results presented in table VI do not show any marked difference in the behavior of the different chlorides tested. The cations, judging from the limited data available, apparently have little or no effect; or at least their chlorides all behave very much in the same manner. In this respect the alkali salts are different in their effect upon the fire-holding capacity of tobacco, for here the salts of caesium, rubidium, and potassium in general are much more favorable to combustion than the corresponding salts of sodium or lithium. A similar contrasting behavior of different cations of chlorides was noted by HARDEN (17), who found that potassium chloride and ammonium chloride cause a definite degree of fermentation in inactivated yeast, while sodium chloride has no effect. He says: "A specific difference in relation to alcoholic fermentation exists between the ions of sodium on the one hand and of potassium and ammonium on the other hand." SCHREINER and SULLIVAN (29) found that potassium salts retard oxidation by the roots of plants.

The effect of the chlorides of the alkalies in retarding oxidase activity suggests a possible practical application in preventing the browning of fruits and vegetables during their preparation for canning, preserving, or drying.

The sulphates apparently increase oxidation slightly in all cases, but the readings are not sufficiently large to be of any positive significance.

The nitrates of potassium, sodium, and magnesium have no marked effect on oxidation, while the nitrates of barium, calcium, manganese, and iron (ferric) decrease it. These results are similar to the effect upon respiration as found by ZALESKI and REINHARD (33). FERNBACH and LANZENBERG (13) and KAYSER (20) find that nitrates increase alcoholic fermentation, but, as they point out, the effect may be to increase multiplication of the yeast cells rather than to affect enzymatic action.

In tables II and III and figs. 2 and 3 are shown the oxidation of pyrogallol by bark alone, by bark and carbonate, and by carbonate alone. From these it is seen that in the last two cases oxidation is considerably greater than that by the bark alone. It is also seen that during the first 3 hours oxidation by carbonate is greater than that by carbonate and bark, but that after the experiment has stood overnight oxidation by healthy bark and carbonate approaches that by carbonate alone, and oxidation by diseased bark and carbonate exceeds it.

The most obvious explanation of this fact, although possibly not the true one, is that oxidation by a carbonate is a strictly chemical reaction, catalyzed only by hydroxyl ions, which soon comes to a definite end, while oxidation by carbonate and bark is a reaction catalyzed by both "oxidase" and hydroxyl ions, in which the presence of the hydroxyl ions increases the effectiveness of the "oxidase," which is slow in reaching an end-point.

Table VI shows that tripotassium phosphate increases oxidation of pyrogallol very markedly, both with and without bark. Although no P_H values for this mixture are available, we know the salt is alkaline in reaction, and this effect complicates the matter. With potassium dihydrogen phosphate at 0.10 N concentration a decrease is evident, and at 0.02 N and 0.002 N concentrations a

slight increase in oxidation occurs. The higher hydrogen ion concentration is probably the cause of the slight depression in oxidation of the 0.10 N strength of the salt. The slight increase in oxidation of the lower concentrations suggests that phosphates may increase oxidase activity, but the limited data are inconclusive. It is interesting to note that IWANOFF (19) found that phosphates raise the amount of respiration in living wheat seedlings. ZALESKI and REINHARD (33) found that disodium phosphate increases the output of carbon dioxide from dried ground seeds, and that the monobasic phosphate decreases it because of the acid reaction. These authors also quote from the work of a student, Miss SCHKLOUSKY, who showed that phosphates increase the action of peroxidases, and from work of another student, Miss ROSENBERG, who showed that phosphates stimulate the catalase activity of different seeds.

In the case of salts of organic acids and the carbonates, all more alkaline than any of the inorganic salts (table VI), oxidation is greater at all stages of the experiment when bark is used than when it is not. Examples of this are shown in table IV. The effect of the salt is not merely additive, however, either here or in the case of the carbonates, as is shown by the following:

OXIDATION OF PYROGALLOL BY BARK AND SALT

	Tested separately (cm. of mercury rise)	Tested together (cm. of mercury rise)
K_2CO_3	4.46	2.70
K tartrate	2.15	1.53
Na oxalate	1.88	1.60

Evidently when bark and salt are combined, there is some factor at work which brings about a slower rate of oxidation than might be expected. What this factor may be we have no means of knowing as yet. Possibly it is the partial neutralization of the hydroxyl ions of the salt by the acid of the bark.

The question why salts vary so widely in the effect they have on oxidation is not easily answered. If we consider only the results with 0.1 N solutions, it seems clear, in the case of the carbonates, potassium dihydrogen phosphate, and the salts of organic acids here reported, that increased oxidation in their presence is due to the excess of hydroxyl ions they furnish; that is, by the

reaction (P_H) their solutions establish when mixed with bark and pyrogallol (table VII). The reaction established by the chlorides, however, can hardly be responsible for the decrease in oxidation they bring about, since sulphates, giving about the same reaction, cause a small increase in oxidation. For example, a mixture of potassium chloride, bark, and pyrogallol has a P_H of 5.19 and gives only 63 per cent as much oxidation as the control. A similar mixture containing potassium sulphate has a P_H of 5.13 and gives 7 per cent more oxidation than the control. The corresponding figures for manganese are: manganese chloride mixture, $P_H = 4.50$, oxidation = 104 per cent of the control.

The situation for nitrates shows several irregularities. Potassium nitrate giving a P_H of 5.14 has practically no effect on oxidation. Magnesium nitrate is also without effect, but gives a P_H of 4.62. The nitrates of calcium, barium, and manganese inhibit oxidation, but manganese gives a lower P_H and the other two a higher one than that given by magnesium nitrate.

The results presented justify the conclusion that when 0.1 N solutions of the salts are used, other ions than hydrogen and hydroxyl play an important part in controlling oxidation. When hydrogen or hydroxyl ions are neutralized in making oxidase activity determinations, therefore, it is important to take into consideration the possible effect of the salts formed thereby. This must be considered as merely preliminary to the real investigations of the relation of specific ions to the oxidation processes in plants and animals. The effect of iron and manganese salts has long been known, but more work is necessary, both with these and with the more commonly occurring chlorides, sulphates, and nitrates of other cations.

Summary

1. One-tenth normal solutions of all of the chlorides tested (potassium, sodium, lithium, caesium, ammonium, calcium, manganese, ferric) decreased oxidation of pyrogallol by apple bark powder.

2. Oxidation was increased very slightly by 0.10 N solutions of all the sulphates tested.

3. Potassium, sodium, and magnesium nitrates (0.10 N) had practically no effect on oxidation, while nitrates of calcium, barium, manganese, and iron (ferric) decreased it.

4. Potassium chloride (0.02 N and 0.002 N) had no effect on oxidation, while manganese chloride in these concentrations increased it.

5. Tartrates, oxalates, citrates, acetates, and carbonates increased oxidation. Marked increase in oxidation in these cases seems to be due, in part at least, to the low acidity of the mixtures of bark, pyrogallol, and salt.

6. Marked decrease in oxidation is not necessarily accompanied by high acidity of the mixtures.

7. Ions other than the hydrogen and hydroxyl may be important in regulating oxidase activity.

8. In neutralizing hydrogen or hydroxyl ions, it is important to take into consideration, in the study of oxidase activity, the possible effect of the salts formed thereby.

9. The chlorides which retard the combustion of tobacco at high temperatures also retard the oxidase action at low temperatures.

10. The effect of the alkali chlorides upon oxidase activity suggests a practical application in preventing the browning of fruits and vegetables during their preparation for canning, preserving, or drying.

The authors wish to express their appreciation to Dr. WM. CROCKER for many helpful suggestions.

BUREAU OF PLANT INDUSTRY
WASHINGTON, D.C.

LITERATURE CITED

1. ABDERHALDEN, EMIL, and GUGGENHEIM, MARKUS, Versuche über die Wirkung der Tyrosinase aus *Russula delica* auf Tyrosin, tyrosinhaltige Polypeptide und einige andere Verbindungen unter verschiedenen Bedingungen. Zeit. Physiol. Chem. **54**:331–353. 1907–1908.

2. ALSBERG, CARL L., Beiträge zur Kenntnis der Guajak-Reaktion. Arch. Exp. Path. und Pharm. Festschrift. Schmiedeberg, pp. 39–53. 1908.

3. Aso, K., On oxidizing enzymes in the vegetable body. Bull. Coll. Agric. Imp. Univ. Tokyo **5**:207–235. 1902.
4. Bach, A., Zur Theorie der Oxydasewirkung. II. Einflusz der Metallsalze auf die weitere Umwandlung der Produkte der Oxydasewirkung. Ber. Deutsch. Chem. Gesells. **43**:366–370. 1910.
5. Bertrand, G., Sur l'action oxydante des sels manganeux et sur la constitution chimique des oxydases. Compt. Rend. **124**:1355–1358. 1897.
6. ———, Sur l'intervention du manganese dans les oxydations provoquèes par la laccase. Bull. Soc. Chim. **17**:619–624. 1897.
7. Buchner, Edward, Buchner Hans, and Hahn, Martin, Die Zymasegärung. Munchen und Berlin. 1903.
8. Bunzell, H. H., A simplified and inexpensive oxidase apparatus. Jour. Biol. Chem. **17**:409–411. 1914.
9. ———, The relationship existing between the oxidase activity of plant juices and their hydrogen ion concentrations, with a note on the cause of oxidase activity in plant tissues. Jour. Biol. Chem. **28**:315–333. 1916.
10. Cole, S. W., Contributions to our knowledge of the action of enzymes. I. The influence of electrolytes on the action of amylolytic ferments. Jour. Physiol. **30**:202–220. 1903.
11. Ewart, A. J., A comparative study of oxidation by catalysts of organic and inorganic origin. Proc. Roy. Soc. London B **88**:284–320. 1914.
12. Falk, I. S., The influence of certain salts on enzyme action. Jour. Biol. Chem. **36**:229–247. 1918.
13. Fernbach, A., and Lanzenberg, A., De l'action des nitrates dans la fermentation alcoolique. Compt. Rend. **151**:726–729. 1910.
14. Gerber, C., La lipase des latex, comparaison avec celle des graines. VI. Action des sels neutres, des elements halogenes et de l'eau oxygenée sur la saponification du jaune d'oeuf par la lipsae du latex d'*Euphorbia Characias*. Compt. Rend. Soc. Biol. **76**:136–141. 1914.
15. Gessard, M. C., Sur la tyrosinase. Compt. Rend. **130**:1327–1330. 1900.
16. Hamsik, A., Zur Kenntnis der Pankreaslipase. Zeit. Physiol. Chem. **71**:238–251. 1911.
17. Harden, Arthur, The condition of activation of washed zymin and the specific function of certain cations in alcoholic fermentation. Biochem. Jour. **11**:64–70. 1917.
18. Hawkins, Lon A., The effect of certain chlorides singly and combined in pairs on the activity of malt diastase. Bot. Gaz. **55**:265–285. 1913.
19. Iwanoff, L., Zur Frage nach der Oxydation der Gärungsprodukte des Zymins beim Atmungsprozess. Biochem. Zeit. **29**:347–349. 1910.
20. Kayser, M. E., Influence des nitrates sur les ferments alcooliques. Compt. Rend. **151**:816–817. 1910.
21. Kraybill, Henry R., Effect of some alkali salts upon fire-holding capacity of tobacco. Bot. Gaz. **64**:42–56. 1917.

22. KÜBEL, F., Über die Einwirkung verschiedener chemischer Stoffe auf die Thätigkeit des Mundspeichels. Archiv. für die gesammte Physiologie **76**:276–305. 1899.
23. LISBONNE, MARCEL, Influence des chlorures et des phosphates sur la saccharification de l'amidon demineralise par les amylases salivaire et pancreatique. Compt. Rend. Soc. Biol. **70**:207–209. 1911.
24. LOEVENHART, A. S., and PEIRCE, G., The inhibiting effect of sodium fluoride on the action of lipase. Jour. Biol. Chem. **2**:397–413. 1907.
25. NASSE, OTTO, Untersuchungen über die ungenformten Fermente. Pflügers Archiv. **11**:138–166. 1875.
26. PORODKO, T., Zur Kenntnis der pflanzlichen Oxydasen. Beih. Bot. Centralbl. **16**:1–10. 1904.
27. REED, G. B., The relation of oxidase reactions to changes in hydrogen ion concentration. Jour. Biol. Chem. **27**:299–302. 1916.
28. ROSE, D. H., Blister canker of apple trees: a physiological and chemical study. BOT. GAZ. **67**:105–146. 1919.
29. SCHREINER, O., and SULLIVAN, M. X., Concurrent oxidation and reduction by roots. BOT. GAZ. **51**:273–285. 1911.
30. TERROINE, E. F., Zur Kenntnis der Fettspaltung durch Pankreassaft. Biochem. Zeit. **23**:404–462. 1910.
31. WOHLGEMUTH, J., Untersuchunger über die Diastasen. I. Die tierischen Diastasen. Biochem. Zeitschr. **9**:10–43. 1908.
32. WOLFF, M. J., Sur quelques proprietes nouvelles des oxydases de *Russula delica*. Compt. Rend. **148**:500–502. 1909.
33. ZALESKI, W., and REINHARD, A., Zur Frage der Wirkung der Salze auf die atmung der Pflanzen und auf die Atmungsenzyme. Biochem. Zeitschr. **27**:450–473. 1910.

PIT-CLOSING MEMBRANE IN OPHIOGLOSSACEAE

GERTRUDE WRIGHT

(WITH PLATES XI, XII AND SIX FIGURES)

The members of the Ophioglossaceae, an isolated family of uncertain origin, are forms with a few large leaves, simple to decompound, and short, slow growing, underground stems, vertical, oblique, or horizontal in position, with crowded fleshy roots. The leaves, which are divided into sterile and fertile lobes, bear on the latter homosporous sporangia.

Of the three widely distributed genera, *Helminthostachys*, a monotypic genus, is the most restricted, occurring throughout tropical Asia to North Australia and New Caledonia. *Ophioglossum* is represented by about 30 species growing under various conditions of moisture and shade in the temperate and tropical zones of both the Eastern and Western hemispheres. *Botrychium*, with nearly as many species, is world wide in its distribution, but is confined chiefly to the temperate regions.

The forms considered in this paper are *Helminthostachys zeylanica*, *Ophioglossum vulgatum*, the only species of the genus native to Canada, and *Botrychium obliquum*, one of the 6 or 8 forms found in Ontario.

The rhizome of *Ophioglossum vulgatum* consists of a large, starch-filled cortex surrounding a siphonostele of endarch bundles of primary wood. This cylinder may be broken by leaf gaps, often so prolonged as to overlap, producing a circle of bundles. Fig. 1 shows several such bundles, one, beside an outgoing root, starting on its way through the cortex to the petiole. There is no endodermis in the mature plant, and the pith is directly continuous with the cortex through the large leaf gaps.

Helminthostachys, whose rhizome is horizontal and dorsiventral, presents a slightly different appearance in cross-section. Fig. 2 shows its broad woody cylinder solid on the lower side, broken

on the upper right by a relatively small leaf gap beside an outgoing leaf trace. The wood is entirely primary, with groups of parenchyma scattered throughout it. The mesarch structure of the bundles is not evident here, but may be demonstrated by means of longitudinal sections. The large-celled, winding endodermis is, unfortunately, too faintly stained to show clearly in the figure. According to FARMER and FREEMAN (4), there is in this form cork formation confined to the upper surface and originating at the bases of the cast-off leaves.

The most extraordinary member of the group in regard to its wood structure, however, is undoubtedly *Botrychium*. In this form there occurs a well developed cylinder of secondary wood, as well as a definite cork layer. The stem shown in transverse section (fig. 3) illustrates this. The woody cylinder surrounding a rather large starchy pith is solid with the exception of small leaf gaps, one of which appears in the lower part of the figure to the left of a horizontal root. The wood, which is composed of tracheids of irregular size, is traversed by numerous uniseriate medullary rays of slightly radially elongated parenchyma. The few and inconspicuous primary bundles are endarch. The pericycle consists of several rows of parenchyma, and is surrounded by an endodermis, frequently multiple. A rather large cortex, also utilized in the storage of starch, is bounded by cork which is visible in the upper right-hand corner of the figure.

The roots of the three genera show no secondary wood of any account. BOODLE (2) has described the addition of a few tracheids at the base of the old roots of *Ophioglossum vulgatum* and *Botrychium Lunaria*, but the later formed parts show only typically primary bundles, in the case of the former genus monarch in structure, and in the latter triarch or tetrarch (figs. 4, 5). The hexarch stele of the *Helminthostachys* root also shows only primary arrangement (fig. 6).

The character of the wood elements themselves in the three genera differs almost as much as their arrangement. Fig. 7 shows the elements in the metaxylem of the root of *O. vulgatum*, stained with Haidenhain's iron-haematoxylin and safranin. They do not differ from those of the stem, hence they represent the general

condition, pitting of the bordered scalariform type. With this stain the primary wall shows broad and black through the secondary, dividing the narrow red borders of adjacent pits. This is most apparent in the upper half of the tracheid to the left, where the scalariform openings are uniseriate, extending from side to side of the tracheid. In the lower half of the tracheid the primary wall has not been cut. The pit borders are more or less clear, also, about the middle of the tracheid to the right where the pits are small, oval, and biseriate. A combination of silver nitrate solution and ammonia, used with a counter stain of methylene blue, demarked these borders most clearly, but, unfortunately, did not lend itself to photography.

On the other hand, the metaxylem of *Helminthostachys* and the metaxylem and secondary wood of *Botrychium* exhibit a much greater differentiation. The tracheids, as seen in longitudinal section, are irregular and frequently nodular in appearance, with pitting distributed equally on their radial and tangential walls. The section illustrated in fig. 14 is from the rhizome of *B. obliquum*, cut tangentially and stained with haematoxylin and safranin. The tracheids are irregular in size and position, and interspersed with uniseriate medullary rays. The central tracheid shows the typical pitting of the secondary wall. The uniseriate and biseriate pits are large, round to oval in shape, with a centrally placed round pore. The small shaded area surrounding the pore is lignified.[1] In the tracheids to right and left is depicted a feature characteristic of both *Botrychium* and *Helminthostachys*, a tertiary wall of lignin. About the center of the tracheid to the left this layer appears as reticulately arranged bars lying over the pitted secondary wall. Above the center the plane of section is lower, exposing only the secondary wall; below the center it is through the lumen of the tracheid, and consequently the tertiary layer is seen in section. In the tracheid to the right, both the tertiary and secondary walls have been cut only in section. Fig. 15, also from *B. obliquum*, gives a sectional view of the pits with their overlaid scalariform. The pit cavities are approximately twice as

[1] In all the text figures lignification has been indicated by means of shading, and a different focus or an obscure feature by dotted lines.

long as broad, and rounded at the ends. The spools between, forming their borders, show a fairly thick, secondary, unlignified wall, ridged in most cases by one to two lignified (shaded) bars. The areas between the pits are small, and the primary wall which traverses them has, frequently, at the edges of the pits, thickenings

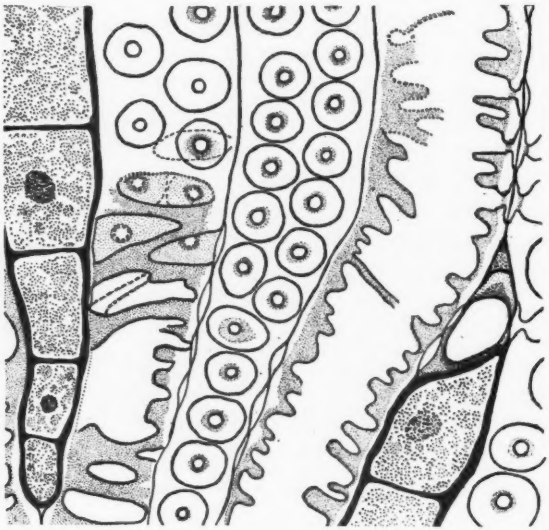


FIG. 14.—*Botrychium obliquum:* tangential section of the rhizome showing pitting; ×600.

similar to bars of Sanio. These are shown on the last four spools toward the top of the figure.

Fig. 10 shows the stem wood of *Helminthostachys* to be fairly similar to that of *Botrychium*, as seen in figs. 14 and 15. To the right of the center the walls of two adjacent tracheids have been cut in section. The left-hand wall is composed of only the secondary layer, which is characteristically thinner than in *Botrychium*, that to the right, of tertiary bars as well. The reticulate arrangement of these bars may be seen in the tracheid to the left of the center. The first-formed elements of the metaxylem of both *Helminthostachys* and *Botrychium* show less of a tertiary layer than the later

formed ones figured here. The scalariform bars in the former are fine and rather far apart, in the latter broader and joined in such a way as to produce the reticulate effect of fig. 10.

In both *Botrychium* and *Helminthostachys* the tracheids of the root wood, although slightly smaller and more regular than those of the stem, resemble these very closely. There is, perhaps, a greater amount of open scalariform tertiary thickening than in the stem and less of the broad, close formation. The petiole wood of both forms is also a likeness in miniature of that of the stem, particularly of the first-formed elements of the primary metaxylem of the latter. Frequently, however, the pit pores in *Helminthostachys* petiole are long and oblique rather than round.

The presence or absence of a pit-closing membrane in the Ophioglossaceae, as in all the vascular cryptogams, has been a matter of dispute. RUSSOW (7), in illustrating his article of 1872, expressed the prevailing view of the anatomists of his time with regard to the vascular cryptogams in general, when he showed no membrane in the pits of either the side or the end walls of *Botrychium*. It was in the following year that SANIO, working with *Pinus sylvestris*, demonstrated beyond a doubt the presence, in the mature condition in that form, not only of a membrane but also of a torus. From that time the pendulum of opinion began to swing in the opposite direction. In response to the stimulus of SANIO'S discovery, evidence has steadily accumulated that the membrane in the vascular cryptogams remains in the pits of the mature wood, not only in the side walls of the elements but, with few exceptions, in the end walls as well. In 1908, however, this view was challenged by GWYNNE-VAUGHAN (5). In returning to the idea of the earliest investigators, that the membrane disappears through resorption in the mature wood, the author distinguishes two types of ferns, represented by *Pteris* and *Osmunda* respectively. Ferns of the *Pteris* type, he claims, lose their limiting membrane only from the pit cavities, while those of the *Osmunda* type lose it also from between the walls of adjacent tracheids in the region between the pits. GWYNNE-VAUGHAN describes a further modification of this type which, however, need not be discussed here, as he classes the Ophioglossaceae with ferns of the *Pteris*

type. As far as the longitudinal walls are concerned, the opposite view, that of the persistence of the membrane in the pits, was upheld by HALFT (6) for "all the vascular cryptogams." He demonstrated by physical and microchemical means the presence of a limiting membrane in both the side and end walls of a large number of ferns. HALFT'S work was verified in the following year by Miss BANCROFT (1), then a research scholar in the University College of Nottingham. Judging from her very lucid paper and my own results with members of this group, I should think that HALFT had shown the "real" nature of the elements in the ferns. Comprehensive as is his work, however, his statement is more so, for no mention is made of a study of any member of the Ophioglossaceae. Miss BANCROFT, also, in corroborating his work, omits this family.

Fig. 16, a drawing from the rhizome of *Ophioglossum vulgatum*, shows the typical membrane in that form. In sections stained with silver nitrate and ammonia and counterstained with methylene blue, the open scalariform pits, with their narrow, pale greenish-blue borders, are traversed by a uniform pale brown membrane. Fig. 17 illustrates the same condition in the root. Here haematoxylin accentuates the broad primary wall within the spools, and stains only faintly the membrane in the pit. The latter, indeed, often appears to be somewhat lignified, taking to a certain extent the red stain of the lignified pit borders. The petiole as it leaves the rhizome exhibits a similar type of membrane.

It was with the greatest difficulty that the membrane in *Helminthostachys* was stained sufficiently for clear demonstration. After prolonged staining with the ordinary haematoxylin and safranin solutions, it remained so vague that its presence only, but not its form, could be ascertained. The latter was finally revealed by a stain consisting of malachite green, Martius' gelb, and acid fuchsin, originally used by Dr. PIANEZE for cancer tissue. The stain was recommended by R. E. VAUGHAN (Ann. Mo. Bot. Gard. May, 1914) as a differential stain for fungus and host cells.

Fig. 18 shows the condition in the rhizome. The lignified (shaded) areas appear bright green, bounding the red of the unlignified secondary walls, which in turn bound the more deeply

stained red primary wall. In the pits, the unlignified membrane, a pale red, assumes the form of a long spindle-shaped torus. Fig. 8 is from the adult metaxylem of the root, and shows the torus lying across the space where the walls of adjacent tracheids have been torn apart in sectioning. A slight thickening was found also in the first-formed elements of the metaxylem. The tracheids

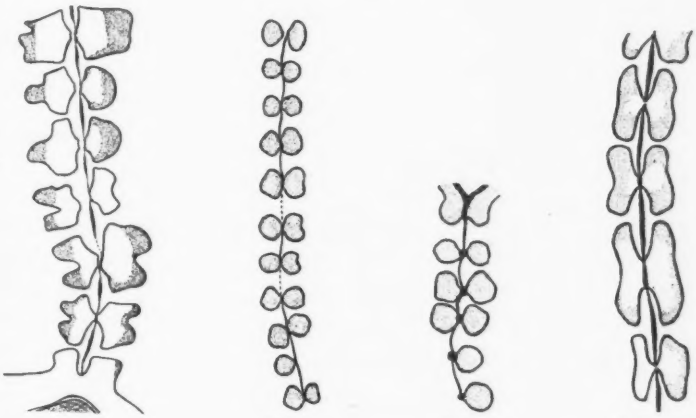


FIG. 15 FIG. 16 FIG. 17 FIG. 18

FIGS. 15–18.—Fig. 15, *Botrychium obliquum:* radial section of the rhizome showing pitting and torus; ×1200; fig. 16, *Ophioglossum vulgatum:* rhizome showing pit membrane; ×900; fig. 17, *Ophioglossum vulgatum:* root in cortex showing membrane; ×900; fig. 18, *Helminthostachys zeylanica:* longitudinal section of rhizome showing torus; ×900.

of the petiole in longitudinal section, however, show a fine uniform membrane with only occasionally a slight thinning toward the edges of the pit.

In *Botrychium* two types of torus occur. The most common type is that seen in figs. 9 and 15, a long, slender, and rather variable spindle. This is found in the mature wood of the stem, the root, and the leaf trace in the cortex. In the last region the membrane varies from a spindle to a uniform line, as seen in transverse section in fig. 11. The arrow in a tracheid to the right of the center points to a fairly thick membrane of the uniform type.

Fig. 12 shows a number of the spindle-shaped ones at a higher magnification. The pit pores have been outlined for greater clearness. The second type of torus occurs in the immature wood of the stem and occasionally in the root. Fig. 19 shows a transverse section from the cambial region of a young rhizome of *B. obliquum*. The tracheids are only slightly lignified, some still showing the contents. Here the torus is a short oval structure as

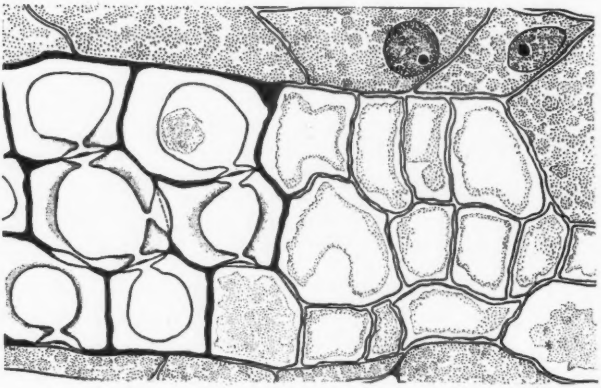


FIG. 19.—*Botrychium obliquum:* transverse section of young rhizome, at cambium showing torus and double membrane; ×600.

long as, or slightly longer than, the pore of the pit, and connected to its edges by a fine membrane.

This section (fig. 19) also illustrates a feature which I have observed in other forms, that is, the double nature of the membrane. In the pit of the tracheid at the left-hand lower corner the membrane is of a double character. The tracheid lies against a parenchyma cell of the ray, and only the half of the membrane next to the wood cell has been thickened, while that lying next to the ray cell remains uniform. The same double nature and plano-convex thickening of the membrane are shown in the third cell to the right. Here a tracheid, as yet unlignified and filled with contents, is adjacent to one which is more advanced in development, and the thickening occurs only on the side of the latter.

A peculiar condition is occasionally met with in the stem. The tracheids are more or less discolored when cut, and stain in a peculiar manner. With Pianeze's stain the membrane becomes yellow. It is usually uniform in thickness, but swollen, occasionally almost entirely filling the pit (fig. 13).

In the petiole of *Botrychium*, as in that of *Helminthostachys*, a uniform membrane prevails. With the exception of the petiole, therefore, and peculiar unnatural spots in the stem, the typical pit membrane in *Botrychium* has a torus.

Thus the only torus I have found among the cryptogams occurs in forms whose pits are broad-bordered and circular or oval in shape. STRASBURGER (8) makes the statement that a torus occurs in *Pteris aquilina*, but he neither enlarges on the statement nor illustrates it. DEBARY (3) describes and pictures for *Pteris* an almost imperceptible one-sided swelling of the membrane, lying to one side of the pit and acting, he states, as a lid to the pit pore. I have searched in vain for such a torus. Frequently the membrane may have a "kink" toward the pit pore simulating the appearance of a torus, but both its edges follow the curve to an equal extent, thus precluding the possibility of a thickening at that point. In *Pteris* the membrane in the pits between tracheid and tracheid invariably remains uniform in thickness. As has been shown in *Botrychium*, a plano-convex torus such as DEBARY describes may occur in the pits of a tracheid where it touches a ray cell. In *Pteris*, however, the membrane even in this region remains consistently uniform. *Equisetum*, *Psilotum*, and *Isoetes*, forms with narrow-bordered pits of the scalariform type, and a number of ferns (including *Ophioglossum*), with the same type of pitting, all show a definitely uniform membrane. In *Helminthostachys* and *Botrychium*, whose pits are circular, broad-bordered, and round-pored, there is developed a definite torus. Although this suggests a possible relation of the torus to the form of the pit, the question of its relationship, whether structural, ecological, or phylogenetic, is one on which it is hoped more light may be thrown after a study of the nature and occurrence of the torus in the other groups of the plant kingdom. It is interesting to note, however, that the form of the torus in *Botrychium*

and *Helminthostachys*, whose pitting is strikingly similar to that of the seed plants, resembles closely the type I have found in the lower gymnosperms, in *Ginkgo* and the araucarians, forms which are to be described later.

To Professor R. B. THOMSON, under whose direction this work has been carried on, is due my grateful acknowledgment of his invaluable assistance and advice. I am indebted also to both Professor THOMSON and Professor J. H. FAULL for material, some of which was obtained originally through the kindness of the Director of the Royal Botanic Gardens, Kew.

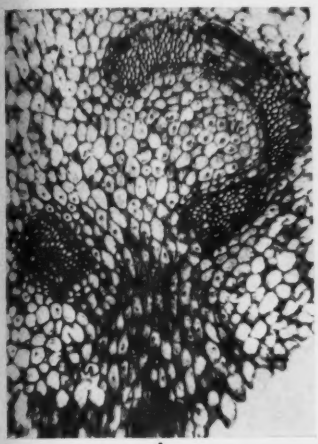
UNIVERSITY OF TORONTO

LITERATURE CITED

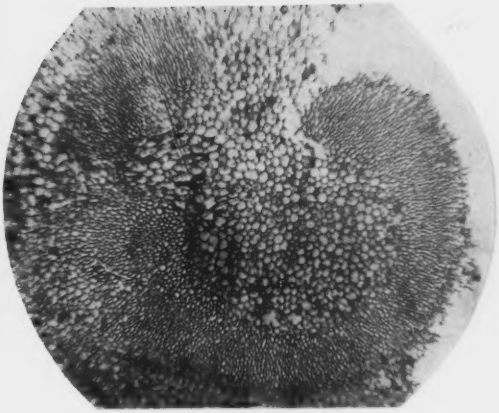
1. BANCROFT, N., On the xylem elements of the Pteridophyta. Ann. Botany 25:745–758. 1911.
2. BOODLE, L. A., On some points in the anatomy of the Ophioglossaceae. Ann. Botany 13:377–394. 1899.
3. DEBARY, A., Comparative anatomy of the Phanerogams and Ferns, pp. 161–162. 1884.
4. FARMER, J. B., and FREEMAN, W. G., On the structure and affinities of *Helminthostachys zeylanica*. Ann. Botany 13:421–445. 1899.
5. GWYNNE-VAUGHAN, D. T., On the real nature of the tracheae in ferns. Ann. Botany 22:517–523. 1908.
6. HALFT, F., Die Schlieszhaut der Hoftüpfel im Xylem der Gefäszkryptogamen. Dissertation. 1910.
7. RUSSOW, E., Vergleichende Untersuchungen. Mem. Acad. Imp. Sci. Saint Petersbourg 19:1–207. 1872.
8. STRASBURGER, E., Das Botanische Practicum, p. 249. 1897.

EXPLANATION OF PLATES XI, XII

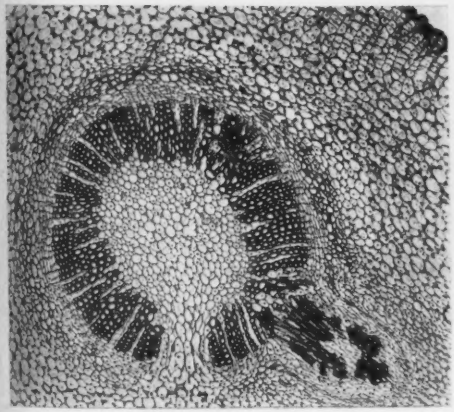
FIG. 1.—*Ophioglossum vulgatum:* transverse section of rhizome; ×50.
FIG. 2.—*Helminthostachys zeylanica:* transverse section of rhizome; ×35.
FIG. 3.—*Botrychium virginianum:* transverse section of rhizome; ×40.
FIG. 4.—*Ophioglossum vulgatum:* root, transverse section; ×210.
FIG. 5.—*Botrychium virginianum:* root, transverse section; ×120.
FIG. 6.—*Helminthostachys zeylanica:* root, transverse section; ×140.
FIG. 7.—*Ophioglossum vulgatum:* metaxylem of root showing pitting; ×875.



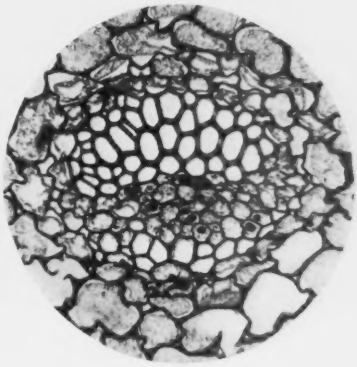
1



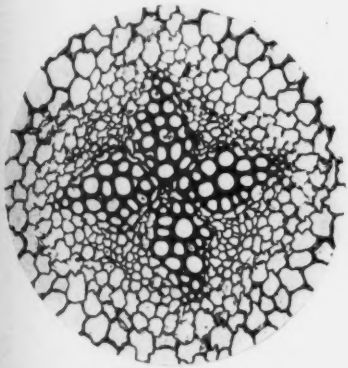
2



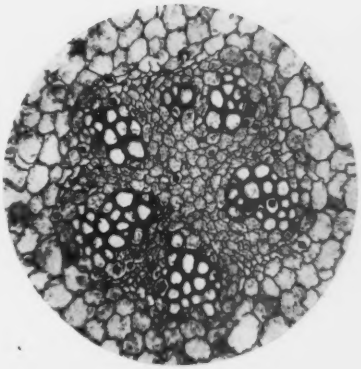
3



4



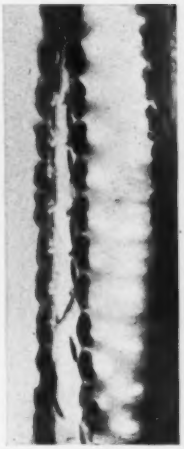
5



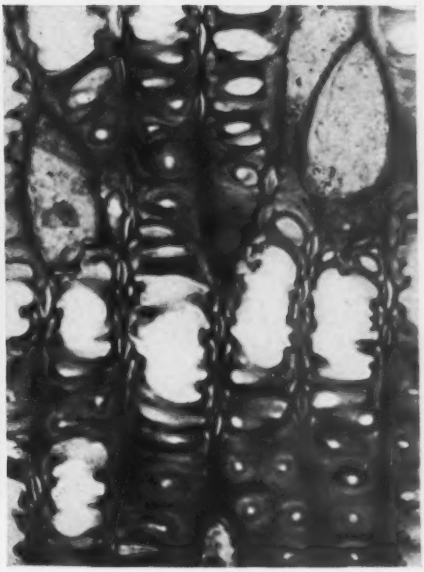
6

WRIGHT on OPHIOGLOSSACEAE

7

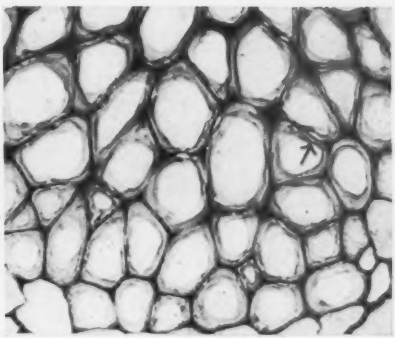


8

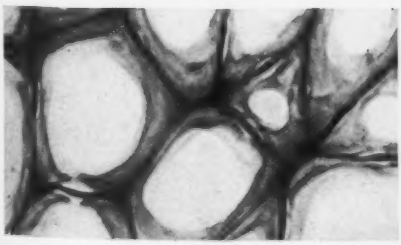


9

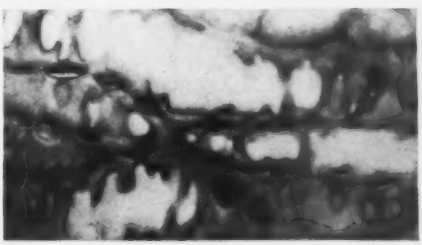
10



11



12



13

WRIGHT on OPHIOGLOSSACEAE

FIG. 8.—*Helminthostachys zeylanica:* metaxylem of root showing torus; ×900.

FIG. 9.—*Botrychium obliquum:* rhizome, tangential section showing torus; ×735.

FIG. 10.—*Helminthostachys zeylanica:* rhizome, longitudinal section; ×825.

FIG. 11.—*Botrychium obliquum:* transverse section of leaf trace in cortex; ×425.

FIG. 12.—*Botrychium obliquum:* part of fig. 11 more highly magnified; ×875.

FIG. 13.—*Botrychium obliquum:* tangential section of rhizome showing thickened membrane; ×525.

DOTHIDIACEOUS AND OTHER PORTO RICAN FUNGI

F. L. STEVENS

(WITH PLATES XIII, XIV AND THREE FIGURES)

The following fungi were collected by the author in Porto Rico, and specimens are deposited in the herbarium of the University of Illinois, and of the New York Botanical Garden. The limitations accepted for the Dothidiaceous genera are those of THEISSEN and SYDOW,[1] which seem to be well founded and wholly tenable.

Dothideales

DOTHIDEACEAE

AUERSWALDIA CECROPIAE P. Henn. (figs. 4, 5).

On *Cecropia peltata:* El Alto de le Bandera, 9043; Mayaguez, 3931; Maricao, 8965; Rio Arecibo, 7798; Florida Adentro, 7756, 2475; Jayuya, 361; Añasco, 3581; Utuado, 6064.

This fungus, as the number of collections shows, is abundant in Porto Rico. From descriptions it seems to be the one just named. It is very variable in habit, especially with age, and there is some doubt as to its generic position. In young specimens there is no stroma, and the fungus appears Sphaeriaceous. In older specimens the stroma is well developed, and the fungus is clearly Dothidiaceous. No colored spores were seen, and the fungus to all appearances is really a *Phyllachorella.* Type material of *A. Cecropiae* P. Henn. and *Physalospora Cecropiae* Rehm are needed before a satisfactory decision can be made.

Uleodothis Pteridis, sp. nov. (figs. 6, 7).—Spots tan-colored, dead, 3–5 mm. across. Stromata black, rugose with perithecia, 1–2 mm. across, conspicuous above, less so below, slightly raised above the leaf surface, orginating sub-epidermally but eventually occupying the whole mesophyll, the upper surface rough and raised, without clypeus, and remaining covered by fragments of the epidermis. Hyphae of the stroma of general parallel arrangement. Loculi many, about 100 μ in diameter, globular. Asci

[1] Ann. Mycol. 13:149. 1915.

numerous, 65×14 μ, cylindrical, 4-spored. Paraphyses few, inconspicuous, fine, filamentous. Spores hyaline, 2-celled, oblong, 17–20×4–5 μ.

On *Pteridium caudatum*, Maricao, 4814 (type), 167.

This fungus agrees somewhat closely with *Dothidella pteridophila* Speg., but differs essentially in that it has paraphyses, and the asci are 4-spored. It differs from *Uleodothis*, as described, in having 4-spored asci, but it does not seem wise to found a new genus merely on this character.

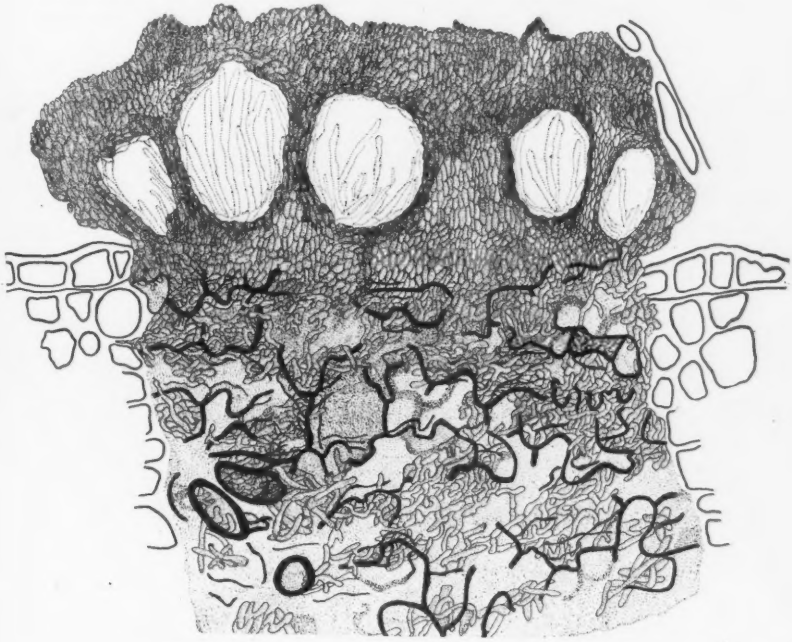


FIG. 1.—Structure of stroma and arrangement of locules

Dothidella portoricensis, sp. nov. (figs. 8, 9; text fig. 1).—Spots linear, 0.5–1×3–4 mm., amphigenous, definite. Stromata linear, entirely occupying the spots, raised above the leaf surface about 70 μ. Perithecial cavities in about 5 rows, nearly globular, about 70 μ in diameter. Paraphyses none. Asci numerous, cylindrical, 54×10 μ, 8-spored. Spores hyaline to dilute smoky, 1-septate, 17×3.5 μ.

On *Gleichenia*, Las Marias, 3551, x.62 (type).

The stromata differ essentially in shape from those of *D. pteridophila* Speg. (fig. 1).

Dothidella flava, sp. nov. (text figs. 2, 3).—Stromata pale to yellow, circular when young, linear when old; when mature, 1600 μ long by 270 μ wide, subepidermal, later erumpent, rising to considerable height above the leaf surface. Perithecial locules

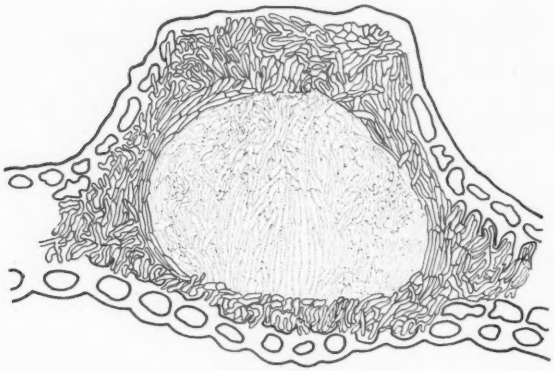


FIG. 2

FIG. 3

FIGS. 2, 3.—Cross and long sections showing arrangement and shape of perithecia and pycnidia.

globular, 60–70 μ in diameter, arranged in one or two rows in the stromata. Asci linear, 8-spored, 34–51×6 μ. Spores hyaline, 2-celled, oblong, 3.5×14 μ. Conidia filamentous, 40×1.5 μ, hyaline, borne in the same stromata with the perithecia and preceding them, and either free in an acervulus or in pycnidial locules in the stroma.

On *Lithachne pauciflora:* Trujillo Alto, 9394, 7654; Mayaguez, 1062, 7432; Florida Adentro, 7665 (type), 7650.

This fungus is particularly interesting. To the naked eye it is like a rust. Superficial microscopic examination shows circular conidial sori which on casual observation might pass as a *Cylindrosporium*. Intermingled with the circular sori are many linear sori and stromata, all bearing the *Septoria*-like conidia in great numbers. Microtome sections show that all development is subepidermal. The first locule is conidiiferous, and is so thin-walled that the wall might easily be overlooked. The sorus elongates, ruptures the epidermis, and in section takes on the appearance shown in text fig. 3. At about this period perithecial locules appear with asci and spores. Their walls are indistinguishable from the surrounding stroma, and the whole structure is truly Dothideaceous. The stromata are frequently overgrown by a *Helminthosporium*. Following THEISSEN and SYDOW, it belongs to the Dothideaceae, falling in the genus *Dothidella*. To many it may appear more reasonable to put it in the Hypocreales on account of its color; but it appears to me to show much closer relationship with the Dothideaceae, notwithstanding its pale color.

PHYLLACHORACEAE

TRABUTIA RANDIAE (Rehm.) Th. and Syd.

On *Randia aculeata*, Cabo Rojo, 6455.

This fungus is clearly a *Trabutia* with strictly subcuticular stroma, and as it agrees well with the published description of *T. Randiae*, it is in all probability that species.

SCIRRHIINEAE

Catacauma Ocoteae, sp. nov.—Spots irregularly circular, 0.5–1 cm. or more in diameter, visible from above or below on dead, tan-colored tissue, border indefinite. Stromata circular, numerous, scattered throughout the spot, plane above, strongly rounded below, 1–1.5 mm. in diameter, between the lower epidermis and the mesophyll. Clypeus hypophyllous, rarely epiphyllous, extending slightly beyond the perithecia, very thick (60–110 μ). Locules several, large (about 300 μ in diameter), irregular. Asci 4–8-spored, 85–102×7 μ long, slender, with long sterile base. Spore 1-celled, hyaline, oblong, 14–20×3.5 μ. Paraphyses filiform.

On *Ocotea leucoxylon*, Monte Alegrillo, 4725 (type), 732, 1347.

Entirely distinct from *Phyllachora ocoteicola*, although often upon the same leaf.

Catacauma palmicola, sp. nov. (figs. 10–12).—Stromata conspicuous above, few below, black, shining, oval, 1–6×1–3 mm.,

with rounded surface, scattered and separate or clustered and confluent. Diseased area extending through the leaf, brown below. Stroma developing between epidermis and palisade cells, often 300 μ thick. Locules irregular in shape, often 500–600 μ wide, basal layer hyaline, thin, lateral walls brown, thick; clypeus black, 60–100 μ thick. Asci numerous, 8-spored, saccate, thin-walled. Spores inordinate, cylindrical, 28–43×12–14 μ, hyaline, continuous.

On *Thrinax ponceana*, Vega Baja, 7716 (type).

CATACAUMA URBANIANUM (A. and H.) Th. and Syd. (fig. 13).

On *Calyptranthes Krugii:* El Yunque, 8243; Maricao, 3677, 3740.

What appears to be the same fungus, although usually hypophyllous and showing a concentric arrangement of stromata, occurs on an unknown Myrtaceous host, no. 5766, San German. Another specimen from Monte Alegrillo, 4526, shows the characteristic acervuli and spores, but is mainly conidial. The *Septoria*-like conidia are borne in very large cavities in the stromata. The ascospores in these specimens are slightly longer (17–20 μ) and slightly thinner (5 μ) than called for by description.

Catacaumella Gouaniae, sp. nov. (figs. 14, 15).—Mainly epiphyllous, rarely hypophyllous. Spots barely exceeding the stromata, hardly visible below. Stromata abundant, roughly circular, 2–3 mm. in diameter, raised, wrinkled, shining black, developing between the epidermis and the palisade cells and made up of parallel cells perpendicular to the leaf surface. Loculi large, flat, 500 μ wide, about 150–160 μ deep, single or few in each stroma. Ostiole very large and distinct. Asci thin-walled, irregular, 8-spored, 61–68×10–11 μ, inordinate. Spores hyaline, 1-celled, ovoid or pyriform, irregular, 14–20×10 μ. Paraphyses none.

On *Gouania polygana:* Mayaguez, 3923 (type), 1049; Salinas, 6798; Dos Bocas, 6007, 8092; Maricao, 8953; on *Gouania lupuloides*, Arecibo-Lares road, 7230.

The last specimen shows the stromata smaller and more abundant upon the lower surface than is the case with the other specimens.

Phaeodothopsis Eupatorii, sp. nov. (figs. 16, 17).—Spot not exceeding the clypeus. Stromata numerous, circular, 1–4 mm. in diameter, black, rough with perithecia, almost exclusively epiphyllous; developing first in the epidermis, producing an

extensive clypeus, then developing the stromata between this clypeus and the palisade cells. Loculi globular or lenticulate, 100–250 μ in diameter, 80 μ high, by pressure sometimes pushing into the mesophyll. Asci about 110×17 μ, cylindrical, 8-spored, inordinate. Spores 20×7 μ, 1-septate about one-third the distance from one end, brown when mature. Paraphyses filamentous, branching.

On *Eupatorium portoricense*, Dos Bocas below Utuado, 6866 (type), 6034, 6830, 6437, 6861, 6032, 6537.

The clypeus is strictly epidermal, and under it very numerous loculi develop, each with an ostiole reaching through the clypeus. The occasional pressing of the perithecia into the mesophyll sometimes gives this the appearance of closer relationship to the Phyllachorineae, but its relationship is clearly with the Scirrhiineae.

Halstedia, gen. nov.—Asci borne in a locule in a superficial stroma.

Type *H. Portoricensis*. Named in honor of BYRON D. HALSTED.

Halstedia portoricensis, sp. nov. (figs. 18, 19).—Stromata amphigenous but more abundant and larger above, densely black, 1–4 mm. in diameter, flat, with surface in the older parts corrugated, or sometimes raised in the center, strictly superficial, non-radiate. Perithecia up to 400 μ in diameter, 160 μ from base to top, internal measurements. Asci 8-spored, 68–85×14 μ, cylindrical. Spores oval, continuous, hyaline or pale straw-colored, 17×10 μ.

On *Sideroxylon foetidissimum*, Quebradillos, 9239 (type).

The fungus consists of a densely black stroma which in the center is nearly 200 μ in thickness, thinning at the edges to the thickness of the mycelium. The stroma is flat-topped, the bulging due to the development of the perithecium usually resulting in a downward thrust and displacement of the leaf rather than of the upper layer of the stroma (fig. 16). In some instances the reverse is true, with an upward bulging. Closest search failed to reveal any evidence of penetration of the fungus through the epidermis, or of any mycelium or signs of disease in any of the host cells. There is no ostiole, and the perithecium is poorly developed, if indeed it is more than a locule in the stroma. The fungus shows close kinship with the Dothideales, but cannot be placed in any of the families of that order as characterized by THEISSEN and SYDOW. It differs from typical Perisporiaceae in the absence of a clearly developed perithecium and in possessing a stroma. It forms an interesting transition form between these two groups, and may for the present be regarded as Perisporiaceous.

Perisporiales

PERISPORIACEAE

Dimerina monenses, sp. nov. (fig. 20).—Epiphyllous, rarely hypophyllous, diffuse over the leaf surface. Mycelium superficial, scant, dark, irregular, 3 μ thick with thinner side branches. No hyphopodia, perithecia rough, irregularly spherical, 45–60 μ in diameter, without ostiole, arranged in close clusters of 10 or more on a close dark subicle. Clusters 150–300 μ in diameter. Asci numerous, elliptical, 34×17 μ, obtuse, 8-spored. Spores inordinate, hyaline or very pale-smoky, 13–16×3 μ, obtuse, 2-celled.

On *Jacquinia barbasco*, Mona Island, 6087.

While the spores and asci agree well in size with those of *Dimerina eutricha* and *D. negeriana*, our species does not agree with these forms in other characters. Agreement as to asci and spores is close with *Asterina paupercula* E. and E., but our perithecium is not that of an *Asterina*.

HYSTERIINEAE

Gloniella rubra, sp. nov. (fig. 21).—Perithecia oblong, scattered, numerous, epiphyllous, black, 600–1500×180–250 μ, opening by one or more longitudinal clefts; the perithecial contents thus exposed are red (near color no. 13 of Saccardo's scale). Asci long-cylindrical, very crooked, especially at the tip, 8-spored, 85–92×10 μ, inordinate. Paraphyses numerous, filiform, long. Spores hyaline or very faintly tinted, 1–3, mostly 3-septate, fusoid, 23–26×3 μ.

On *Arthrostylidium multispicatum* Pilg., El Alto de la Bandera, 4363 (type).

This species is somewhat like *G. pusilla* Sacc., but differs from it in its carbonaceous perithecium, red contents, curved asci, etc.

PLEOSPORACEAE

PHYSALOSPORA HOYAE, v. Hohn. (fig. 22).

On *Ficus*, Mona Island, 6234, 6169.

This very pretty form I refer with some hesitancy to the preceding species. The spores in my specimen are uniseriate, and are considerably narrower than the description of VON. HOHNEL calls for. *P. elasticae* Koord. is close kin, but differs in the rounded spores. Pycnidia are present, bearing slender filamentous spores, 7×1 μ.

MYCOSPHAERELLACEAE

Guignardia Justiciae, sp. nov. (figs. 23, 24).—Diseased spot indefinite, finally yellowish and pale, rather evenly beset with perithecia, 1–2 mm. distant from each other. Perithecium globose, completely imbedded in the leaf, 265 μ in diameter and depth, its wall dark, several cells (34 μ) thick. Host tissue surrounding the perithecium hypertrophied to a distance of about 125 μ in every direction from the perithecium. The resulting "gall" is visible from either side of the leaf, and has the superficial appearance of a stroma with a single central perithecium. The ostiole develops late. Paraphyses none. Asci clavate, usually with a long stipe; body of ascus 17–20×61 μ; total length, including stipe, 125 μ. Spores 8, inordinate, hyaline, 1-celled, oval, 9–10×18 μ.

On *Justicia verticillaris:* Maricao, 806 (type); El Yunque, 2839; El Gigante, 8557; El Alto de la Bandera, 9046.

This fungus is noteworthy on account of the peculiar gall-like formation surrounding each perithecium, the thick wall, and the peculiar long-stalked asci.

Guignardia Tetrazygiae, sp. nov.—Spots indefinite, irregular, 1–2 cm. in diameter or occupying the whole leaf, tan-colored, centers studded with the perithecia which are scattered evenly and profusely over the affected areas. Perithecia black, conspicuous both above and below, about 160 μ in diameter, thick-walled. Asci, sporiferous part oval, 45×27 μ, 8-spored, inordinate, stipe long, slender, 30–60×4–5 μ. Paraphyses none. Spores 1-celled, hyaline, oval, obtuse, 24×10 μ.

On *Tetrazygia* sp.: San German, 4567 (type); Vega Alta, 4148.

This differs from *Laestadia melastomalum* (Lev.) Sacc. in the absence of paraphyses, shape of asci, and other characters. The leaf spot is very characteristic.

Guignardia Nectandrae, sp. nov.—Spots indefinite when young, becoming definite as the host tissue dies, then angular, 2–6 mm. in diameter, showing from both sides of the leaf. Perithecia opening on both sides of the leaf, more abundant below, scattered, located in the mesophyll but causing swelling of both leaf surfaces. Perithecia thin-walled, pale, 70–85 μ in diameter,

located deep in the mesophyll. Asci clavate, 100–115×20 μ, 8-spored. Spores hyaline, oval, 21–24×8–10 μ, 2-celled, septa either in the middle or more frequently located near one end.

On *Nectandra coriacea* (?), Quebradillos, 4994 (type).

This fungus is of very distinctive appearance upon the leaves, where the erumpent perithecia so closely simulate a rust in appearance that the author was led to place it with the rusts on mere casual examination.

SPHAERIACEAE

Zignoella algaphila, sp. nov.—Mycelium fine, pale to brown, twining around and penetrating its algal host and turning it brown. Perithecia black, 90×170–180 μ, variously formed but usually bottle-shaped, broadest a little above the base, with a prominent beak about 24 μ in diameter and with the fibers arranged parallel around the ostiole. Surface coarsely reticulate but not hairy; basal portion appearing as though hairy due to adhering remnants of mycelium. Asci numerous, 8-spored, cylindrical, 71×7 μ. Paraphyses fine, threadlike. Spores hyaline, 3-septate, pointed at each end, 17–21×3.5 μ.

On *Cephaleuros virescens* on *Artocarpus incisa*, Mayaguez, 51 (type).

The parasitic alga when alone on this host is yellow or often nearly colorless, but when invaded by the *Zignoella* all the colonies take on a dark hue, giving the whole leaf much the appearance of being mildly affected with sooty mold. The genus *Zignoella* is large and composed mainly of wood-inhabiting saprophytes. One is listed on *Valsa*, one on the thallus of *Castagnia*, and two species (*Z. enormis* Pat. and *Z. cubensis* H. and Pat.) on the alga *Stypocaule*. These thallus-inhabiting forms, however, are markedly different from the present species.

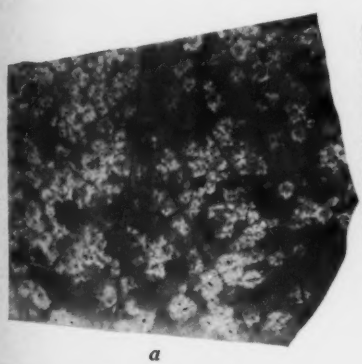
Sphaeropsoidales

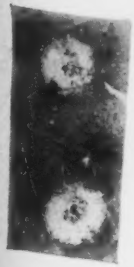
Phyllosticta bonduc, sp. nov.—Spots indefinite, large, starting usually at edge or apex and progressing over the whole leaflet. Pycnidia numerous, black, scattered, ostiolate, about 160–190 μ in diameter. Wall about 17 μ thick, ostiole large, irregular. Conidiophores simple, hyaline, arising from sides and base of the pycnidium. Conidia hyaline, 1-celled, oblong, 21×4 μ, somewhat irregular in shape.

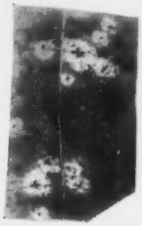
On *Caesalpinia bonduc*, Guanica, 360 (type).

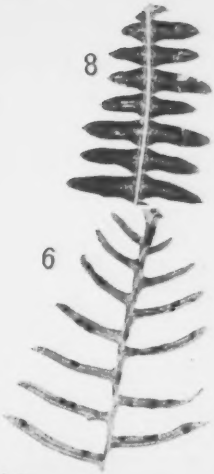
This fungus is quite distinct from *Phyllosticta guanicensis*.

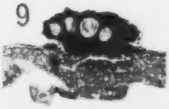
UNIVERSITY OF ILLINOIS
URBANA, ILLINOIS

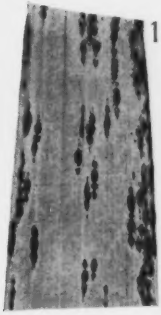


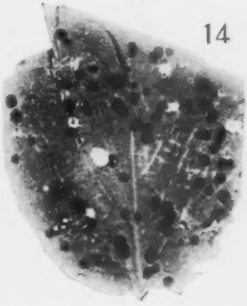


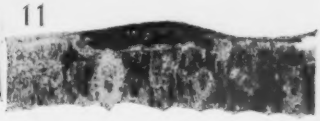




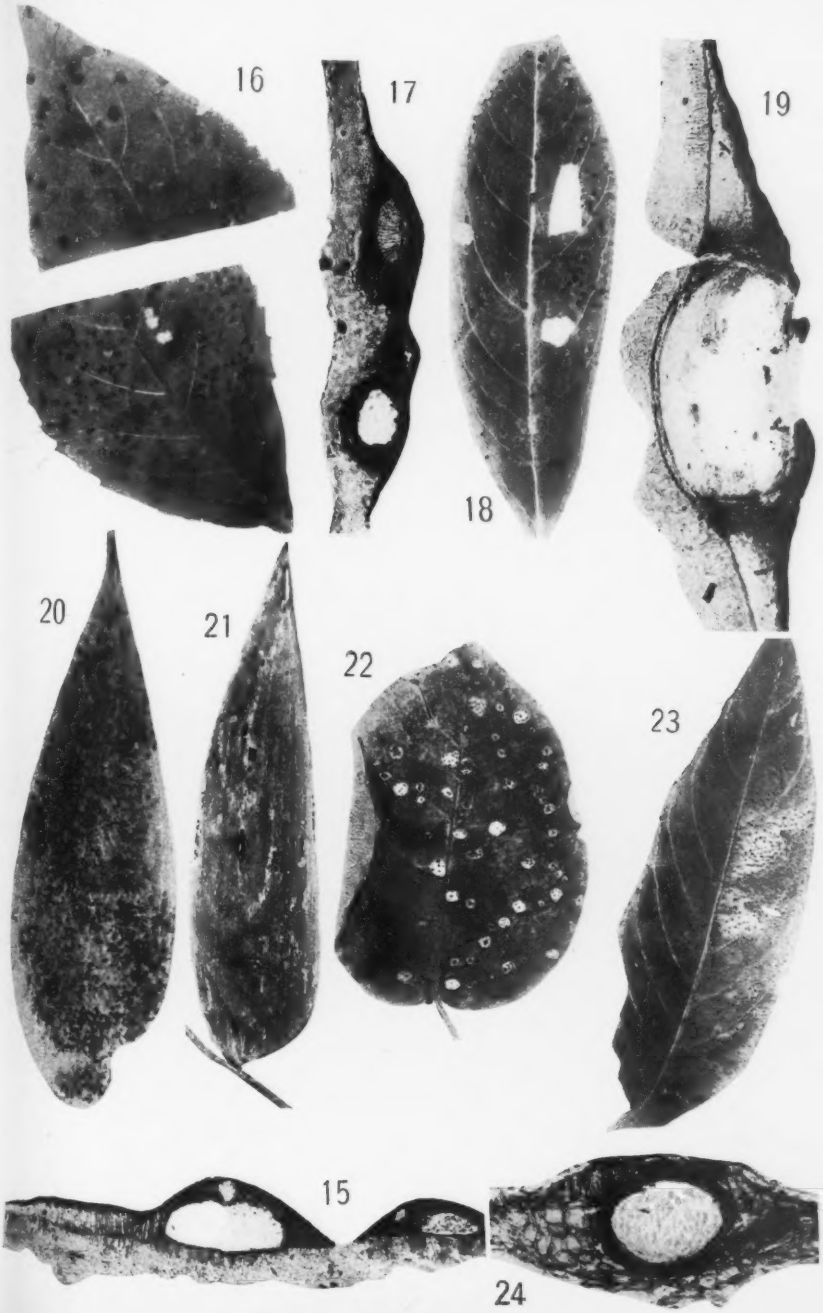








STEVENS on FUNGI



STEVENS on FUNGI

EXPLANATION OF PLATES XIII, XIV

FIGS. 4, 5.—*Auerswaldia Cecropiae* P. Henn.: fig. 4, habit, showing, *a*, abundant scattered young spots (no. 361); *b*, older infections each surrounded by discolored spot (no. 9043); *c*, still older spots with much dead tissue (no. 9043); fig. 5, old, well-developed stroma completely occupying leaf from epidermis to epidermis.

FIGS. 6, 7.—*Uleodothis Pteridis*, sp. nov.: fig. 6, habit; fig. 7, stromata occupying whole mesophyll with locules on both surfaces.

FIGS. 8, 9.—*Dothidella portoricensis*, sp. nov.: fig. 8, habit, a leaf segment (no. ×62); fig. 9, cross-section of stroma.

FIGS. 10–12.—*Catacauma palmicola*, sp. nov.: fig. 10, habit; numerous stromata on piece of palm leaf; fig. 11, young stroma and locules, showing that it is strictly subcuticular; fig. 12, sectional view of mature stroma showing 3 locules.

FIG. 13.—*Catacauma urbanianum* (A. and H.) Th. and Syd., showing habit, no. 3577.

FIG. 14.—*Catacaumella Gouaniae*, sp. nov.: habit, stromata scattered over leaf.

FIG. 15.—*Catacaumella Gouaniae*, sp. nov.: stroma in section, showing that it is formed entirely above palisade cells.

FIGS. 16, 17.—*Phaeodothiopsis eupatorii*, sp. nov.: fig. 16, habit, showing stromata of different ages (no. 6866); fig. 17, stroma in cross-section.

FIGS. 18, 19.—*Halstedia portoricensis*, sp. nov.: fig. 18, general view of stromata on leaf; fig. 19, stroma in section, showing depression of leaf by growth of stroma.

FIG. 20.—*Dimerina monensis*, sp. nov., showing habit.

FIG. 21.—*Gloniella rubra*, sp. nov., showing habit.

FIG. 22.—*Physalospora Hoyae* v. Hohn., showing habit.

FIGS. 23, 24.—*Guignardia Justiciae*, sp. nov.: fig. 23, habit; fig. 24, section through hypertrophied portion showing perithecium.

SPERMATOGENESIS IN BLASIA

LESTER W. SHARP

WITH PLATE XV

Introduction

The following brief account of spermatogenesis in *Blasia pusilla* is based upon preparations made from a limited amount of material collected near Chicago several years ago. The preparations, which were originally made for use in classes, proved upon careful examination to show with admirable clearness all stages included in the last spermatogenous mitosis and the transformation of the androcyte (spermatid) into the spermatozoid. Since the results of the examination differ in two important points from those reported by WOODBURN (12) in the only previous paper dealing with these features in *Blasia*, they are here recorded.

Description

The description will begin with the spermatogenous cells of the penultimate generation, the androcyte mother cells, to use the terminology of ALLEN (1). The cells of the earlier generations (androgones) have been examined, and nothing which it is safe to call centrosomes has been observed. Unfortunately, however, the material did not show many androgone nuclei in division; anaphases were present, but metaphases, where centrosomes are usually most conspicuous if present at all, were not found. No conclusive statement can be made, therefore, regarding the presence or absence of centrosomes in the androgones.

In the androcyte mother cell, before the stage represented in fig. 1, the cytoplasm has an almost homogeneous appearance, and included in it are several granules or vaguely defined areas. In some cells these granules, from 1 to 6 or more in a thin section, may appear to be all alike; while in other cells one or two of them may be more sharply defined and more deeply stained than the others. It is possible that of these several granules two survive as the

centrosomes shown in fig. 1, after the manner of the "black granules" in the body cell of *Dioon* (CHAMBERLAIN 4). On the other hand, it would be possible to select a series of cells illustrating the divergence of daughter centrosomes arising by the division of one, as in *Equisetum* (SHARP 8); or even to show the origin of the bodies in question from the nucleus, as described by WILSON (10) for *Atrichum* and *Mnium*. The writer, however, believes that the evidence afforded by his material is insufficient to support any of these hypotheses in the case of *Blasia*. The present description, therefore, will begin with a stage (fig. 1) at which the identity of the centrosomes is unmistakable, the question of their origin and earlier history being left an open one.

Two centrosomes, whatever may be their previous relation to other cell granules, soon stand out with great distinctness as intensely staining bodies near the cell membrane at opposite poles of the androcyte mother cell (fig. 1). At this time the cell is still rather square in section, since it has only begun to round off from its neighbors, and the centrosomes commonly occupy the corners, as shown in the figure. From each centrosome a conical group of very faint fibers extends toward the nucleus, which is somewhat flattened on the sides facing the centrosomes. While the nucleus is undergoing the prophasic changes (fig. 2) these fibers become more plainly visible, and when the nuclear membrane disappears they become attached to the chromosomes and establish the achromatic figure with the centrosomes at its poles.

It is at metaphase that the spindle is seen most clearly (fig. 3). As noted by WOODBURN (12), it may lie either straight or obliquely in the cell. Furthermore, the cells may round up and alter considerably in shape while mitosis is in progress, so that although the centrosomes may at first be situated near the corners of the cell, all appearance of the diagonal division so characteristic of many bryophytes may in many cases be lost by the time the metaphase and succeeding stages are reached (figs. 4, 5).

When the chromosomes reach the poles at the end of the anaphase (fig. 4), they usually come in contact with the centrosomes. As a result the latter, which are very minute, are often difficult to find at this stage. Careful search, however, reveals cells in which

they stand out clearly a little apart from the chromosome groups. From this time onward they become increasingly distinct. As the membranes form about the reorganizing daughter nuclei at telophase the centrosomes are left just outside in the cytoplasm (fig. 5), and while cell division is being completed they move away from the nucleus and take up positions nearer the cell membrane (fig. 6).

The two androcytes (spermatids), between which no cell wall is laid down, quickly round off from each other (fig. 7). In probably the majority of cases they are somewhat triangular in shape, owing to the usual diagonal plane of the division which differentiates them. In each androcyte the blepharoplast, as we may call the centrosome in view of the function it performs in the cell which it now occupies, enlarges considerably and becomes somewhat elongated.

A careful search has been made in the cytoplasm of the androcytes for accessory structures corresponding to the "chromatoider Nebenkörper" (IKENO **6**) or "limosphere" (WILSON **10**), the "percnosome" and the "apical body" (ALLEN **2**) described by other investigators of bryophyte spermatogenesis; but, as WOODBURN (**12**) also reports, nothing which can confidently be regarded as such a body has been found. Occasionally there is observed in the cytoplasm a darker area, which, although it is as a rule rather vague in outline (fig. 7, below and at left of nucleus in each cell), may in certain cases be more definitely delimited (fig. 9). A similar appearance is also often seen in the later stages of spermatogenesis (figs. 15, 16, 18, 19). It may well be that we are dealing here with a limosphere or other accessory body, but without more trustworthy evidence for its constant presence and regularity in behavior, at present it does not seem advisable to attribute to this body any special significance in the case of *Blasia*. The cytoplasm of the androcyte frequently contains a large vacuole, which may or may not lie near the blepharoplast (fig. 8).

The blepharoplast now begins to undergo a series of transformations which ultimately result in the formation of the cilia-bearing thread of the spermatozoid. After elongating very slightly, as previously noted, the blepharoplast becomes constricted

(fig. 10, upper cell) and divides by a process of simple fission into two portions (fig. 10, lower cell). These two portions, or blepharoplast granules as they may be termed, often lie very close together, but in many cases they are so far apart that there can be no doubt that the fission is complete. As a rule one of the granules at once begins to elongate, while the other remains relatively unchanged, so that many cells show two bodies, one of them round and the other comma-shaped, lying close together near the cell membrane (fig. 11). At about this stage the granules usually move closer to the nucleus. The comma-shaped granule continues to elongate (fig. 12) and divides again; whether the other granule also divides or not is a difficult matter to determine. The granules continue to multiply by fission (fig. 13) until several are present in a row (figs. 14, 15); seven was the largest number counted with certainty. The granules now appear less distinct from one another; it seems that they gradually undergo a coalescence (figs. 14–16), but it may also be that some of the fissions are incomplete, some of the granules therefore never being entirely separate.

The nucleus at this time moves more closely against the beaded blepharoplast (fig. 15) and begins to draw out into a point by the side of the latter (fig. 16). Both nucleus and blepharoplast continue to elongate spirally, the association between them becoming constantly more intimate (fig. 17). Fig. 18 represents a cell like that of fig. 17 viewed from the direction indicated by the arrow; it is here seen that the blepharoplast is applied along one edge of the flattened point of the nucleus. As the transformation continues the boundary between nucleus and blepharoplast gradually becomes indistinguishable (fig. 19). Even at this late stage the irregular outline of the blepharoplast is still evident; the blepharoplast granules have not yet become so completely coalesced that the thread which they form is smooth in outline. The nucleus continues to elongate and condense, becoming increasingly slender, while two cilia grow out from the blepharoplast, which projects beyond the nucleus at the anterior end. The spermatozoid is now mature (fig. 20) and ready to escape from the antheridium.

Discussion

The two main points wherein this description disagrees with that of WOODBURN (12) are as follows. First, according to that author there are no indications of centrosomes in the spermatogenous mitoses, the blepharoplast first appearing as a cytoplasmic differentiation in the androcyte. On the contrary, the present writer finds that centrosomes are present at all stages of the last mitosis, and that these persist as the blepharoplasts of the androcytes. Second, WOODBURN states that the blepharoplast in the androcyte undergoes a simple elongation to form the cilia-bearing thread, whereas the present writer sees it fragmenting to several pieces which coalesce to form the thread somewhat after the manner of the blepharoplasts of *Equisetum* and *Marsilia* (SHARP 8, 9).

It is not improbable that this disagreement is due in part to actual differences in the two lots of material studied. Although the single species of the genus, *Blasia pusilla*, was used in both instances, a comparison will show that the cells described in the present account are little more than half the size of those figured by WOODBURN. Although it is possible, therefore, that the two lots of material represent two varieties, too much weight should not be placed upon a size difference, for it is known in certain cases (*Equisetum*, SHARP 8) that androcytes and spermatozoids often vary considerably in size in the same lot of material.

Lack of agreement as to the presence of centrosomes during mitosis is perhaps not surprising. Because of their extreme minuteness the centrosomes might easily be overlooked in the stages previous to that at which WOODBURN first finds them, and at which they enlarge and become really conspicuous for the first time. With regard to the fragmentation of the blepharoplast, on the other hand, it is more difficult to understand why material actually the same should be interpreted so differently. In the writer's material the process of fragmentation is shown with great clearness; only occasionally is anything found in good preparations which might be interpreted as a uniformly elongating blepharoplast. Moreover, in no case has a condition approaching that shown in WOODBURN'S fig. 11 been observed. The nucleus becomes closely applied to the blepharoplast when the latter is in

the form of a short lumpy rod or series of granules, and at no time does the blepharoplast have the form of a long slender thread free from the nucleus as in WOODBURN'S figure. The writer, therefore, is inclined to attribute the disagreement for the most part to actual differences in the material studied rather than to differences in interpretation.

The phenomenon of fragmentation is probably the most interesting feature of the blepharoplast of *Blasia*. In all previous accounts of bryophyte spermatogenesis, including those of IKENO (6) on *Marchantia*, WILSON (10) on *Pellia*, *Polytrichum*, and *Atrichum*, WOODBURN (11, 12, 13) on several liverworts and *Mnium*, Miss BLACK (3) on *Riccia*, and ALLEN (2) on *Polytrichum*, the blepharoplast is reported to elongate without breaking up into smaller portions. ALLEN (2) states that "while the possibility of a somewhat similar occurrence [fragmentation] is suggested by the rather knotty appearance of the blepharoplast of *Polytrichum* when it begins to elongate, there is no time when it is visibly resolved into smaller bodies." In *Blasia*, therefore, we have the only known instance in bryophytes of such a fragmentation of the blepharoplast as occurs in *Equisetum*, *Marsilia*, and the cycads.

Although fragmentation is in general a characteristic of the blepharoplasts of the cycads, and only occasionally found in pteridophytes (*Equisetum* and *Marsilia*), it is now evident that it may occur in forms lower in the scale. Moreover, it is seen that it is not, as might be supposed, merely a means by which large blepharoplasts become transformed, for the blepharoplasts of *Equisetum* and *Marsilia*, and especially those of *Blasia*, are very small. Although the details of the process of fragmentation differ in the various cases (by simple fission in *Blasia* and by vacuolization in the other forms), it is scarcely to be doubted that the phenomenon is a result of similar causes in all. In attempting to find a possible historical reason for it, one is struck by the resemblance between the fission of the blepharoplast in *Blasia* (fig. 10) and the division of an ordinary centrosome before mitosis. If the blepharoplast actually represents a centrosome, as the writer (8) believes the evidence indicates, it is at least possible that its frequent fragmentation, in spite of the fact that in the more advanced

forms (cycads) this fragmentation becomes a very much modified process, may be a manifestation of the power of division which is one of the chief characteristics of centrosomes. According to this interpretation the first fission of the blepharoplast of *Blasia* (fig. 10) would correspond to the centrosome division which would normally occur if another mitosis were to take place, and the further fragmentation would represent a further manifestation of the centrosome's power of division which may have been retained from a time when more spermatozoids were produced from a mother cell, and which has in some way become a feature of the development of the cilia-bearing structures. In this way *Blasia* may shed light upon the origin of the remarkable behavior of the cycad blepharoplasts.

To this idea, which presents itself as a suggestion and may scarcely deserve to be proposed as a theory, there are obviously many objections. Chief among these is the fact that fragmentation is most conspicuous in the blepharoplasts of the cycads, but developed almost not at all in those of the bryophytes, which would be expected to have retained in the manner of their elongation more evidences of a derivation from normal centrosome division. It is possible, however, that the simple fission of the blepharoplast as seen in *Blasia* was soon replaced in most bryophytes and pteridophytes by uniform elongation without fragmentation through the failure of the fission to occur after the slight elongation normally preceding it (figs. 7–9), this elongation then continuing to form the uniform cilia-bearing thread. Fragmentation would thus be a retained feature in *Blasia*, *Equisetum*, *Marsilia*, and the cycads, although the manner in which it is accomplished in the higher forms (through a complex process of vacuolization rather than simple fission) would still be regarded as an advanced feature subsequently evolved. Whether, therefore, the objection stated rules out the suggested explanation or not can scarcely be decided in view of the fact that the evidence at hand has been obtained from so few bryophytes and pteridophytes, comparatively speaking, and especially in view of our lack of adequate knowledge of blepharoplast origin and behavior in the algae.

A further objection may be seen in the case of animal spermatogenesis, in which an undoubted centrosome elongates without fragmentation as it performs its rôle in the development of the motor structures. It is noteworthy, however, that cilia are frequently seen growing from recently divided centrosomes in the case of certain insect spermatocytes (HENNEGUY **5**) in much the same fashion that the cilia start to grow from the recently formed blepharoplast granules in *Equisetum* (SHARP **8**). Moreover, in the Flagellata, which should furnish evidence more valuable than that in the higher animals, it is known that in certain cases blepharoplasts arise from functional centrosomes by division (see MINCHIN **7**, pp. 82 ff.).

Although there is thus seen to be considerable evidence for the derivation of blepharoplast fragmentation from normal centrosome division, this evidence is probably best regarded as scarcely sufficient to warrant the establishment of such an interpretation as a general theory.

The question of the relation of the centrosome to the blepharoplast has been fully discussed by the writer in his papers on *Equisetum* and *Marsilia* (**8, 9**). It will be sufficient here to recall that the conclusions were reached that the blepharoplasts of bryophytes, pteridophytes, and gymnosperms are "ontogenetically or phylogenetically centrosomes" (IKENO); that these centrosomes become more and more restricted in the life history in passing upward through these groups; that they are retained in spermatogenous cells because of the biological importance of the cilia-bearing function which they there perform; and that in connection with this function they have become profoundly modified, losing many of the characteristics of centrosomes and assuming new characteristics not exhibited by centrosomes elsewhere.

To these conclusions *Blasia* furnishes support of no new kind; it merely confirms them by affording another example of blepharoplasts arising from centrosomes functional in mitosis. How extensive this centrosome behavior is in the case of *Blasia* the present study may not show, for, as stated in the description, the writer's material does not enable him to say whether the bodies in question arise from preexisting ones by division or not, or whether they

are present at only one or more than one spermatogenous mitosis. So far as actual evidence goes, it is possible to state unreservedly only that they are present from the stage represented in fig. 1 onward, and that through the single mitosis they appear to perform the usual functions of centrosomes. The discovery of fragmentation in the blepharoplast of a bryophyte serves to confirm the view that the blepharoplasts of all groups above the algae are homologous structures, and the details of the process aid materially in accounting for the behavior of those blepharoplasts which have become least centrosome-like.

Summary

1. Centrosomes are present in *Blasia* at all stages of the mitosis which differentiates the androcytes, and in the androcytes they persist and function as the blepharoplasts.

2. In the transformation of the androcyte into the spermatozoid, the blepharoplast fragments repeatedly by simple fission, forming a number of distinct granules which coalesce to form a short lumpy rod. This rod elongates and becomes a more uniform thread bearing two cilia, while the nucleus also elongates in intimate union with it to form the body of the spermatozoid. The present instance is the first in which blepharoplast fragmentation has been reported in a bryophyte.

3. It is possible that the fission of the *Blasia* blepharoplast, and therefore the more complex fragmentation of the blepharoplasts of *Equisetum*, *Marsilia*, and the cycads, may be homologized with the normal division exhibited by ordinary centrosomes.

CORNELL UNIVERSITY

LITERATURE CITED

1. ALLEN, C. E., Cell structure, growth, and division in the antheridia of *Polytrichum juniperinum* Willd. Archiv Zellforschung **8**:121-188. *pls. 6–9*. 1912.

2. ———, The spermatogenesis of *Polytrichum juniperinum*. Ann. Botany **31**:269–292. *pls. 15, 16*. 1917.

3. BLACK, CAROLINE A., The morphology of *Riccia Frostii* Aust. Ann. Botany **27**:511–532. *pls. 37, 38*. 1913.

4. CHAMBERLAIN, C. J., Spermatogenesis in *Dioon edule*. BOT. GAZ. **47**: 215–236. *pls. 16–18*. 1909.
5. HENNEGUY, L. F., Sur les rapports des cils vibratiles avec les centrosomes. Arch. d'Anat. Micr. **1**:481–496. *figs. 5*. 1898.
6. IKENO, S., Die Spermatogenese von *Marchantia polymorpha*. Beih. Bot. Centralbl. **15**:65–88. *pl. 3*. 1903.
7. MINCHIN, E. A., An introduction to the study of the Protozoa. London. 1912.
8. SHARP, L. W., Spermatogenesis in *Equisetum*. BOT. GAZ. **54**:89–119. *pls. 7, 8*. 1912.
9. ———, Spermatogenesis in *Marsilia*. BOT. GAZ. **58**:419–431. *pls. 33, 34*. 1914.
10. WILSON, M., Spermatogenesis in the Bryophyta. Ann. Botany **25**:415–457. *pls. 37, 38. figs. 3*. 1911.
11. WOODBURN, W. L., Spermatogenesis in certain Hepaticae. Ann. Botany **25**:299–313. *pl. 25*. 1911.
12. ———, Spermatogenesis in *Blasia pusilla*. Ann. Botany **27**:93–101. *pl. 11*. 1913.
13. ———, Spermatogenesis in *Mnium affine* var. *ciliaris* (Grev.) C. M. Ann. Botany **29**:441–456. *pl. 21*. 1915.

EXPLANATION OF PLATE XV

All figures were drawn at the level of the table with the aid of an Abbé camera lucida from cells stained with iron alum-haematoxylin. Examination of the cells was made under a Zeiss 2 mm. apochromatic objective, N.A. 1.40, but because of its slightly greater magnifying power a Spencer 2 mm. achromatic objective was used with an 18 ocular for outlining the drawings. The figures, which have not been reduced in reproduction, show a magnification of 4200 diameters.

FIG. 1.—Androcyte mother cell (penultimate spermatogenous cell) with two centrosomes.

FIG. 2.—Prophase of last spermatogenous mitosis; centrosomes at poles of developing spindle.

FIG. 3.—Metaphase; centrosomes at spindle poles.

FIG. 4.—Late anaphase; centrosomes present.

FIG. 5.—Telophase; centrosomes near daughter nuclei.

FIG. 6.—Late telophase; each cell has one centrosome (blephoraplast).

FIG. 7.—Androcytes (spermatids) rounded off; blepharoplast slightly elongated in each; dark body near nucleus.

FIG. 8.—Pair of androcytes with vacuoles in cytoplasm.

FIG. 9.—Androcyte with dark body (limosphere?) in addition to blepharoplast.

FIG. 10.—Pair of androcytes: blepharoplast undergoing fission in upper cell; two blepharoplast granules resulted from fission in lower cell.

FIG. 11.—Pair of androcytes showing elongation of one blepharoplast granule.

FIG. 12.—Androcyte; slightly later stage.

FIG. 13.—Blepharoplast granules multiplying.

FIG. 14.—Later stage; granules somewhat coalesced.

FIG. 15.—Nucleus moving against blepharoplast.

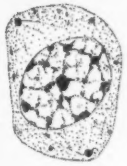
FIG. 16.—Nucleus elongating by side of blepharoplast; blepharoplast granules becoming coalesced.

FIG. 17.—Later stage; blepharoplast and nucleus becoming closely associated.

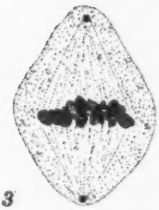
FIG. 18.—Cell like that of fig. 17 viewed from direction indicated by arrow; blepharoplast lying along edge of flattened point of nucleus.

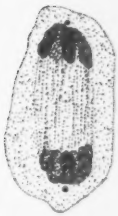
FIG. 19.—Later stage; blepharoplast still irregular in outline; boundary between nucleus and blepharoplast indistinguishable.

FIG. 20.—Mature spermatozoid ready to escape from antheridium.

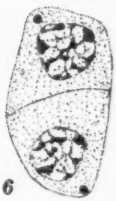

1

2


3


4

5


6

7

8

9

10

11

12

13

14

15

16

17

L. W. Sharp, del.

18

19

20

SHARP on BLASIA

CURRENT LITERATURE

NOTES FOR STUDENTS

Weather and fruitfulness.—DORSEY[1] has done much to place on an experimental basis a subject concerning which there have been many erroneous popular beliefs. In so far as it affects pollination and fertilization, he divides weather into 4 components, rain, temperature, wind, and sunshine. Wind and sunshine in themselves are of minor importance. Rain prevents the dehiscence of the anthers, or causes them to close if they have already dehisced. While this is beneficial in retaining much of the pollen in the anther during a rain, the pollen is not available for pollination during this time. Contrary to popular belief, rain does not cause the pollen to burst, and although the stigmatic fluid may be diluted thereby, this does not seem to be injurious. Some pollen may be washed from the stigma by rain, but an abundance is left for fertilization. Rain does not injure the viability of pollen. Low temperatures retard the growth of the pollen tube, but do not seem to cause delay in the abscission of the style. The stigma is receptive for 4–6 days and then rapidly disintegrates. The style abscisses 8–12 days after bloom. A delay in pollination due to rain, or slow pollen tube growth due to low temperatures, may therefore eliminate fertilization by preventing the pollen tube from passing the point of abscission before the abscission of the style. Applying this analysis of weather to certain years of fruitfulness and to certain other years of non-fruitfulness, it is found that each year there is a definite correlation between the weather and the setting of fruit. The experiments are thus given a practical test.—S. V. EATON.

Determination of biological fluids.—Duggar and Dodge,[2] after discussing some of the difficulties encountered in examining biological fluids, particularly colored plant juices, by the indicator method of H ion determination, describe a new method which they have found satisfactory for the examination of colored plant juices. "The method consisted in simply arranging for each side of the colorimeter a pair of cups slipping to a certain depth one into the other. The method of procedure is then as follows. For the lefthand

[1] DORSEY, M. J., Relation of weather to fruitfulness in the plum. Jour. Agric. Res. **17**:103–126. *pls. 13–15. fig. 1.* 1919.

[2] DUGGAR, B. M., and DODGE, C. W., The use of the colorimeter in the indicator method of H ion determination with biological fluids. Ann. Mo. Bot. Gard. **6**:61–70. 1919.

set, or column, water (or colored standard solution) is used in the outer cup, and the colored test fluid plus the indicator in the inner cup. After adjustment, this set is not removed from the colorimeter during an observation. In the case of the righthand set the outer cup contains the colored test fluid, while the inner cup is for the standard solution plus indicator. This set is placed on the right for convenience, as it may be necessary to compare with the test fluid a series of standards until an exact match is obtained. A rough comparison, of course, is made before selecting the standard solution for comparison. In each case the column must contain an equal depth of colored test solution and of standard or colorless liquid, the indicator being in the standard in the one case and in the test solution in the other. There are no optical difficulties, and unless the indicator combines with the test solution, the comparison may be perfect."

The authors believe this method is as rapid as and more accurate than other methods.—J. WOODARD.

Storied structure of dicotyledonous woods.—A recent paper by Record[3] continues his studies upon the storied or tierlike structure of woods. He finds this arrangement of the secondary elements characteristic of many dicotyledonous woods, occurring through a wide range of orders and families. Such woods on longitudinal section (particularly the tangential) present fine cross lines or striations ("ripple marks"), which may be due to (1) the horizontal seriation of the medullary rays, (2) the tierlike arrangement of the tracheids, wood fibers, vessel segments, and the secondary phloem elements, or (3) a combination of (1) and (2). In some woods the pit areas on the fibers are also in seriation. This storied structure has been found fairly characteristic of the families Leguminosae (40 genera), Bignoniaceae (3), Bombacaceae (3), Compositae (3), Malvaceae (4), Sterculiaceae (7), Tiliaceae (5), and Zygophyllaceae (3); and occurs in one or two genera of each of the following families: Amarantaceae, Ebenaceae, Hippocastanaceae, Moraceae, Sapindaceae, and Ulmaceae.

Particular attention has been given in the present investigation to the various elements storied, the uniformity and distinctness of these transverse lines (ripple marks), and the height of the tiers in each wood examined. "Ripple marks" are sufficiently constant in stems of considerable thickness to serve, the author believes, as a "valuable diagnostic feature."—LADEMA M. LANGDON.

Antarctic and sub-antarctic vegetation.—TURRILL[4] has embodied in a convenient and useful summary the botanical results of the Swedish expedi-

[3] RECORD, S. J., Storied or tierlike structures of certain dicotyledonous woods. Bull. Torr. Bot. Club **46**:253–273. 1919.

[4] TURRILL, W. B., Botanical results of Swedish South American and antarctic expeditions. Roy. Bot. Gard. Kew Bull. 268–279. 1919.

tions to the antarctic regions. These have appeared from time to time in the report of SKOTTSBERG and others, and many have been noted in this journal.[5] The more recent reports have contributed to our knowledge of the vegetation of the portion of South America and adjacent islands between 48° and 56° S. Here is a rain forest covering a limited area, and composed of trees of low stature, few exceeding 10 m. in height. The conspicuous species include *Nothofagus betuloides*, *Drimys Winteri*, *Pseudopanax laetevirens*, and *Libocedrus tetragona*, the only conifer reaching Fuegia. In unforested areas dwarf shrubs, many from the heath family, and cushion plants are conspicuous.

Farther to the north the Valdivian rain forest occupies the region between the coast and the Andes, forming in the lower passes of the mountains a transition to the deciduous forest of the east slope. Between 41° and 44° S. a forest of *Libocedrus chilensis* is interposed between the rain forest and the deciduous. Many other formations are characterized, such as the pampas area east of the Andes, the alpine heaths and meadows, the tussock grass and the tundras. The bibliography includes 23 articles.—GEO. D. FULLER.

Influence of environment on form and structure.—FOLSAM[6] reports a study of the effects of 5 different degrees of soil water supply upon the structural features of *Ranunculus sceleratus* and *R. abortivus*. Plants were grown as pot cultures in a greenhouse. Water was supplied in amounts varying from complete submergence of soil and plant, to only enough soil moisture to support life. In the 24 which were studied 6 structural characteristics of *R. sceleratus* gave consistently larger values with progressively greater water supply in the first generation of plants. In the second generation, 2 of the 6 characters continued to show the same relation. They were (1) thickness of stem cortex, and (2) thickness of stem aerenchyma, both absolute and relative to cortex thickness. In the first generation 5 structural features of *R. abortivus* were found to be related in the same way to water supply. Of these the one relation of increased laminar area of root leaves with increased water supply was shown, although less consistently in the case of *R. sceleratus*. A third generation of the latter species was grown to determine whether the conditions of water supply of parent affected the laminar area of root leaves of progeny grown both as xerophytes and as amphibians. Seeds for this generation were obtained from the xerophyte group of the first generation, and from the amphibious group of the second generation. Progeny grown with a large water supply gave consistently increased laminar area of root leaves over plants grown with a small water supply, regardless of water relations of parents.—J. M. ARTHUR.

---

[5] BOT. GAZ. **58**:96–98, 190. 1914; **63**:423. 1917.

[6] FOLSAM, DONALD, The influence of certain environmental conditions, especially water supply, upon form and structure in *Ranunculus*. Physiol. Res. **2**:209–276. 1918.

Osmotic pressure in the potato.—In an effort to throw some light on the physiological basis of tip-burn, LUTMAN[7] has studied the osmotic pressure of the potato plant throughout a growing season. In the young plant, when the foliage is being formed, the osmotic pressure of the leaves is greater than that of the stems. After the flower buds are formed and the tubers begin to grow, the stalks predominate over the leaves in osmotic pressure. Sugars account for the high pressures of the stalks. Tip-burn begins to appear at this stage. This higher osmotic pressure of the stalks is maintained during the hot weather of July and August. With the coming of cool rainy weather in September and the resumption of growth of the foliage, the leaves again gain the ascendancy. As the plant dies the osmotic pressure decreases, the soluble materials being largely transported to the tubers. The osmotic pressure of the growing tubers is always lower than that of the stems and leaves, although above that of the roots. The study does not explain tip-burn, although the author sees two possible explanations of it: (1) the loss of water from the leaves to the stems, due to the higher osmotic pressure of the latter; and (2) the lack of nourishment of the leaves, due to the translocation of food materials from the leaves to the tubers. The author draws some other theoretical and practical conclusions from his data.—S. V. EATON.

Anatomy of prairie plants.—Selecting the dominant species from some prairie associations, Miss HAYDEN[8] has studied their leaf structure and presented considerable data, the most valuable being in the form of plates from drawings of cross-sections. She concludes that prairie plants show a xerophytic tendency in their leaf structure in the form of specialized palisade tissue, thick-walled epidermis, the presence of water-storing tissue, and sometimes of trichomes.

In studying the subterranean parts of plants from the same habitats, including a larger number from swampy areas, the same author[9] again presents many data in the form of drawings. Her principal conclusions are that in a dry habitat there is a tendency to the production of prominent mechanical tissue and reduction of parenchymatous tissue. In moist habitats, however, parenchymatous tissue is well developed and aerenchyma is abundant in swamp plants. The subterranean stem is predominant in moist lowland regions, and is more efficient than roots in propagation.—GEO. D. FULLER.

---

[7] LUTMAN, B. F., Osmotic pressures in the potato plant at various stages of growth. Amer. Jour. Bot. **6**:181–202. *figs. 2.* 1919.

[8] HAYDEN, ADA, The ecological foliar anatomy of some plants of a prairie province in central Iowa. Amer. Jour. Bot. **6**:69–85. *pls. 10–14.* 1919.

[9] ———, The ecological subterranean anatomy of some plants of a prairie province in central Iowa. Amer. Jour. Bot. **6**:87–105. *pls. 15–28.* 1919.